

FIFRA SCIENTIFIC ADVISORY PANEL (SAP)

OPEN MEETING

POLLINATOR RISK ASSESSMENT FRAMEWORK

DOCKET NUMBER: EPA-HQ-OPP-2012-0543

FIFRA SAP WEB SITE <http://www.epa.gov/scipoly/sap/>

OPP Docket Telephone: (703) 305-5805

UNITED STATES ENVIRONMENTAL

PROTECTION AGENCY

CONFERENCE CENTER LOBBY LEVEL

ONE POTOMAC YARD (SOUTH BUILDING)

2777 SOUTH CRYSTAL DRIVE

ARLINGTON, VIRGINIA 22202

SEPTEMBER 11<sup>TH</sup> - 14<sup>TH</sup>, 2012



1 **FT0"HTGF"LGPMKPU"LT0<** Good morning, again. I  
2 want to welcome everyone to this FIFRA Scientific Advisory Panel  
3 meeting. The topic of this particular meeting is Pollinator  
4 Risk Assessment Framework.

5 My name is Fred Jenkins and I am the  
6 designated Federal Official for this meeting. Before we get  
7 started, I would like to just take a couple of minutes to go  
8 over a few administrative items. As a DFO, I serve as a liaison  
9 between the panel and the Agency and I am responsible for  
10 ensuring that provisions of the Federal Advisory Committee Act  
11 are met.

12 I want to extend my thanks to this entire  
13 panel for agreeing to serve in this meeting, and to the public  
14 as well, for coming to this meeting. The FIFRA SAP is a Federal  
15 Advisory Committee that provides independent scientific peer  
16 review and advice to the Agency on pesticide issues.

17 It is important to note that the panel only  
18 provides the advice and recommendations for EPA, and that all  
19 regulatory decision-making and implementation authority remains  
20 with the Agency. We have worked with appropriate agency  
21 officials to ensure that all appropriate ethics and regulations  
22 are satisfied for this meeting.

23 Panel members have been provided their  
24 provisions of the Federal Conflict of Interest laws, and each  
25 participant has filed a financial disclosure report. I, along  
26 with our deputy ethics officer, and in consultation with the



1 Office of General Counsel, have reviewed these reports to ensure  
2 all ethics requirements are met.

3 This meeting provides an opportunity for  
4 public comment. We have several people who will be speaking  
5 today during the public comment period, which will be tomorrow  
6 morning.

7 Now, if you have not made prior arrangements  
8 with me to make oral comments and you wish to make an oral  
9 comment during the oral comment period tomorrow morning, please  
10 let me know or someone else in our SAP staff, who are all  
11 situation here to my right.

12 Now, without having made prior arrangements,  
13 we ask that you please limit your comments to just five minutes.

14 There is a public docket for this meeting. The docket is  
15 listed on the agenda. All background materials and other  
16 related documents are available in the dockets and slides of EPA  
17 presentations that you will see today. It should be available  
18 now. If not, they will be available very shortly.

19 At this point, I want to introduce and extend  
20 my thanks to Dr. Daniel Schlenk, to my left, who is serving as  
21 Chair to this meeting. He is also the FIFRA SAP Chair.

22 **DR. DANIEL SCHLENK:** Thanks, Fred. Good  
23 morning, everyone. Welcome to this particular panel, the  
24 Pollinator Risk Assessment Framework.

25 At this point in time, it's customary to go  
26 around the room and introduce ourselves. If you wouldn't mind



1 stating your name, where you're from, and sort of your area of  
2 expertise.

3 I'll begin. My name is Dan Schlenk. I am a  
4 professor in the Department of Environmental Sciences at the  
5 University of California, Riverside. My research expertise is  
6 in fate and metabolism of pesticides in aquatic organisms.

7 My job for this particular panel, though, is  
8 to keep us on time and get us through all 52 questions before  
9 Friday so we can get home on the weekend.

10 So with that, I'll turn it over to Martha.

11 **DR. MARTHA SANDY:** Hello. I'm Martha Sandy.  
12 I'm a toxicologist. I'm chief of the toxicology and  
13 epidemiology section of the Office of Environmental Health  
14 Hazard Assessment. That a department within the California  
15 Environmental Protection Agency.

16 **DR. STEPHEN KLAINE:** Good morning. I'm Steve  
17 Klaine. I'm a professor at Clemson University and I'm an  
18 aquatic ecotoxicologist.

19 **DR. JAMES MCMANAMAN:** Good morning. I'm Jim  
20 McManaman. I'm a professor in obstetrics and gynecology at the  
21 University of Colorado. I'm vice-Chair for research there. I'm  
22 a reproductive biologist.

23 **DR. KENNETH DELCLOS:** I'm Barry Delclos -- or  
24 Kenneth Delclos from the FDA's National Center of Toxicological  
25 Research. I'm a toxicologist, primarily in reproductive  
26 toxicology.



1                   **DR. DAVID TARPY:** My name is David Tarpy. I'm  
2 in the Department of Entomology at North Carolina State  
3 University and the Extension Apiculturist for North Carolina.

4                   **DR. PAUL SCHWAB:** My name is Paul Schwab. I'm  
5 an environmental soil chemist at Texas A&M University.

6                   **DR. THOMAS POTTER:** Good morning. I'm Thomas  
7 Potter. I'm a research chemist with the USDA Agricultural  
8 Research Service, Southeast Watershed Laboratory in Tifton,  
9 Georgia. And my area of expertise is pesticide environmental  
10 fate and transport assessment.

11                   **MR. JENS PISTORIUS:** Hello. My name is Jens  
12 Pistorius. I'm from Germany and working at the Julius  
13 KQhn-Institut, in charge of the risk assessment for pesticides  
14 in honey bees for the Investigation Center for Honey Bee Poison  
15 Incident and for the governmental research for Germany for  
16 pesticide abuse.

17                   **DR. JEFF PETTIS:** Good morning. I'm Jeff  
18 Pettis. I'm an entomologist and I'm with the USDA Agricultural  
19 Research Service and I head the bee research lab here, nearby,  
20 in Beltsville, Maryland.

21                   **DR. NANCY OSTIGUY:** My name is Nancy Ostiguy.  
22 I'm in the entomology department at Penn State. I do a research  
23 on honey bee epidemiology.

24                   **DR. ROSALIND JAMES:** My name is Rosalind James  
25 and my expertise is in bee diseases, a bee pathologist, and I'm  
26 director of the Pollinating Insect Research Laboratory in Logan,



Utah, which is part of the USDA Agricultural Research Service.

**DR. GREG HUNT:** Good morning. My name is Greg Hunt. I'm at Perdue University. And my area of research is honey bee genomics and behavior.

**DR. NINA FEFFERMAN:** Hi. I'm Nina Fefferman. I'm here from Rutgers University. My area of research is applied mathematical modeling of ecological and epidemiological systems.

**DR. MAY BERENBAUM:** I'm May Berenbaum, professor and head of the Department of Entomology at the University of Illinois at Urbana-Champaign. And my interest is in insect chemical ecology and chemical mediation with interactions between plants and insects.

**DR. DANIEL SCHLENK:** Thank you, everyone. At this point in time, we're going to get introduction and charge from the director of the Office of Pesticide Programs, and that's Dr. Steven Bradbury.

**DR. STEPHEN BRADBURY:** Thanks, Dr. Schlenk. Welcome, to all members of the panel to join us for this next week and 52 questions.

I think part of the reason you're seeing so many questions is because of the challenge that is before us in undertaking these risk assessments and advancing the science to support our decision-making. I want to give a little bit of background to that before we get started.

First, I want to thank you all for all the



1 work you've already done in reading our White Paper and starting  
2 to put together your thoughts on the charge questions, and also  
3 thank you for your time this week, and then for the month or two  
4 after the meeting when you pull the report together to  
5 contribute to these kinds of peer reviews.

6 Sometimes sitting on the other side of the  
7 table, I know what it takes. So I greatly appreciate your  
8 contribution to the Agency's efforts to advance our scientific  
9 undertaking.

10 I also want to thank Fred and members of the  
11 SAP panel for all the hard work it takes to get one of these  
12 meetings together.

13 The SAP, the Science Advisory Panel, which is  
14 created under the FIFRA statute, is a Federal Advisory Committee  
15 that provides the Agency with expert advice on the science  
16 behind its ecological risk assessments and human health risk  
17 assessments for pesticides. This panel is a very important part  
18 of the day-to-day operation of our business at EPA, which is  
19 undertaking risk assessments for pesticides and making  
20 regulatory decisions. And science is the foundation of that  
21 decision-making process.

22 So this panel is an important component to  
23 that effort, in addition to transparency and openness in public  
24 participation, which is critical to our business and as we go  
25 forward.

26 So I also want to thank members of the public



1 who have already provided information to the docket and for the  
2 comments that will be coming around tomorrow morning because all  
3 that input is very important to your deliberations, as well as  
4 the work that we're undertaking.

5 As you know, this week's effort is all around  
6 assessing risks of pesticides to pollinators, with the primary  
7 focus on honey bees at this stage of our effort. As you know,  
8 the White Paper that's been prepared reflects collaboration with  
9 Health Canada's Pesticide Management regulatory Authority, as  
10 well as California's Department of Pesticide Regulation.

11 And that theme of collaboration, both  
12 internationally and nationally, I want to touch upon as I go  
13 forward with my remarks. Because clearly, the concepts that we  
14 tried to pull together in the White Paper reflect approaches  
15 that have been developed, not only with the folks here, but have  
16 been enhanced and built from national and international  
17 interactions and collaborations with the scientific community  
18 and different countries from around the globe.

19 So clearly, what we're presenting here today  
20 is a reflection of a lot of work, across a lot of organizations  
21 and with a lot of colleagues. Hopefully, we've done justice to  
22 the thoughts and ideas that we've put forward in the document  
23 which you'll be seeing.

24 As you know, pollinator services are critical  
25 to agricultural production and in human food supply and also  
26 just the honey itself, and other products that come out of the



1     hive is an important part of both our nutrition and the commerce  
2     of this country.

3             Also, pollinators have been a critical  
4     component to the viability of the ecosystems with the  
5     pollination that they provide in support of the habitat that we  
6     all depend upon. As we know, from the National Academy of  
7     Sciences report of 2007, and the yearly USDA reports and other  
8     reports around the globe, pollinator decline is a significant  
9     issue that we're facing, not only in the United States, but  
10    globally. And the causes of pollinator decline are quite  
11    complex and clearly not straightforward -everything from habitat  
12    modification, habitat loss, nutritional issues, bee management  
13    practices, and pesticides all being considered as part of the  
14    issues that are facing the viability of honey bees and  
15    pollinators in general.

16            At least, from the best we can tell, there  
17    isn't a single cause, but clearly, all these stressors are  
18    playing some role, to varying degrees, in the decline. Folks in  
19    the EPA, as well as our colleagues in PMRA and Cal DPR are  
20    taking a hard look at issue and working very diligently with  
21    colleagues across the country and the globe in trying to  
22    elucidate what these causes can be and how to reconcile these  
23    effects.

24            It's clearly a complex issue that's going to  
25    take a multi-faceted approach to resolving the issue and working  
26    across lots of different organizations and stakeholders to reach



1 a solution to this problem and try to turn the corner on  
2 pollinator populations.

3 In EPA and the Pesticide Program, our primary  
4 focus in contributing to this global effort is to try to better  
5 understand the role that pesticides may play in pollinator  
6 populations and pollinator viability and to advance our risk  
7 assessment technique so that we can better elucidate cause and  
8 effect and what the potential role of pesticides could be and  
9 pollinator honey bee effects, and thereby inform well-reasoned  
10 regulatory decisions on the safe use of pesticide products.

11 So our niche in this big challenge is to try  
12 to see what we can do to help advance pesticides science and  
13 risk assessment techniques. So consistent with this  
14 international challenge, and not just the national challenge,  
15 we've been working closely with state and federal partners, as  
16 we've been moving forward over the years.

17 I think the White Paper reflects an aspect of  
18 that collaboration and working with Canada and California's  
19 Department of Pesticide Regulation as we pull the White Paper  
20 together and some of the approaches from moving forward.

21 Clearly, the White Paper is also drawing upon  
22 the deliberations and thoughts from the SETAC Pellston Workshop  
23 of a couple of years ago, which brought people together from all  
24 over the world to think through the test data requirements and  
25 different kinds of risk assessment approaches that could brought  
26 to bear.



1           Also, clearly working with our colleagues at  
2 the European Food Safety Authority and getting some of their  
3 insights and concepts. You can see that reflected in the  
4 aspects of the White Paper, as well as working with our  
5 colleagues in OECD, the Organization of Economic Cooperation and  
6 Development, where there is an effort across all the member  
7 countries of OECD to try to develop harmonized approaches and  
8 integrate a variety of techniques and ideas as we go forward.

9           The risk assessment methodology that's  
10 described in the White Paper is strongly founded on US EPA's  
11 ecological risk assessment framework. And that framework is  
12 used, and used quite heavily in the White Paper, but it's used  
13 across the agency in how we formulate our risk assessment and  
14 undertake the risk assessment. Part of the beauty of that  
15 framework is that it helps us integrate not only issues we want  
16 to deal with, in terms of pollinators, but our broader  
17 components of our ecological risk assessment. So that  
18 conceptual model and how it's dealing with fate and transport,  
19 for example, of a pesticide, creates a foundation that's also  
20 being used in other components of the ecosystems and for other  
21 receptors as well.

22           We feel, using a framework, it provides us a  
23 solid foundation for what we can do today and what we can do  
24 tomorrow. Our first goal by using the framework and all of the  
25 various components you'll be hearing about this morning and into  
26 the afternoon is that at the end we want to have a risk



1 characterization component to that risk assessment that provides  
2 a transparent, clear, reasonable, and consistent approach to how  
3 to interpret the potential risk of pesticides to pollinators,  
4 honey bees in particular, in this White Paper.

5 And it's with that clarity of our risk  
6 description and risk estimates that we feel it's critical to  
7 informing our regulatory decisions as we go forward.

8 Now, clearly, there are a lot of uncertainties  
9 that remain and a lot of scientific complexities that aren't  
10 resolved yet today. And our risk assessment process in our  
11 White Paper hopefully captures what we feel we know something  
12 about today, and documents things that we don't know the answers  
13 to yet. But our approach is that we feel that we have credible  
14 science that we can start moving forward today in advancing our  
15 techniques for risk assessments for pollinators, realizing we'll  
16 learn more as we go forward. But hopefully we have a foundation  
17 upon which we can build upon, so that as new insights and new  
18 techniques come to bear, we can fold them into our risk  
19 assessment process.

20 So there's always more to learn. There's  
21 always more that could be done, but our approach is not to wait  
22 for perfection before we can start moving forward and to start  
23 to advance our capabilities for undertaking quantitative risk  
24 assessments for pollinators.

25 So with that, I want to thank EPA and the PMRA  
26 and the California scientists that all contributed to the White



1 Paper. And I'm really looking forward to the next several days  
2 as we go through the presentations and hear your first thoughts  
3 on the charge questions. If it's okay with the Chair, I would  
4 like to turn it over to Dr. Brady for a little more focused  
5 introduction.

6 **DR. DANIEL SCHLENK:** Sure. Before you get  
7 started, Dr. Brady, we have some noise coming in on the phone.  
8 So if you are on the teleconference, can you please mute your  
9 phone? Thank you.

10 Dr. Brady.

11 **DR. DONALD BRADY:** Good morning, members of  
12 the panel. I'm Don Brady. I'm the director at the  
13 Environmental Fate and Effects Division, here in the Office of  
14 Pesticide Programs.

15 Today you will hear presentations designed to  
16 solicit your advice on a proposed tiered process for  
17 quantitatively evaluating the potential risk to pollinators,  
18 utilizing honey bees as surrogates and associated with systemic  
19 and non-systemic pesticides. You will hear an overview of a  
20 proposed tiered process for quantifying the risks of pesticides  
21 to honey bees.

22 As Steve noted, the causes of pollinators are  
23 complex and result from many factors. The advice we request  
24 today is intended to enhance the ability of EPA, the Pest  
25 Management Regulatory Authority in Canada and the California  
26 Department of Pesticide Regulation to reliably screen chemicals



1 for direct and indirect effects on honey bee colonies and on the  
2 usefulness of this framework we present for characterizing  
3 potential effects to other pollinators as well.

4 I want to thank the panel for the work you  
5 will embark on this week. We appreciate and value the advice we  
6 receive from this panel. At the outset, I would also like to  
7 acknowledge the efforts of the team that prepared this White  
8 Paper and presentations. They have worked long and hard to  
9 prepare these materials for you. And I want to acknowledge, in  
10 addition, the work of Dr. Moshameem (ph), who led the work of  
11 the team.

12 Finally, I'm pleased to be here this morning  
13 with my colleagues, Mary Mitchell, from Pest Management  
14 Regulatory Agency of Canada, and Richard Bireley, of the  
15 California Department of Pesticide Regulation, as we initiate  
16 this consultation.

17 Their presence today and their participation  
18 in the development of the White Paper and the presentations is  
19 indicative of the high degree of scientific collaboration among  
20 the agencies during the preparation of the White Paper and the  
21 presentation and of the Agency's shared commitment to bring the  
22 best possible science to bear on this issue. I would like to  
23 pass it to Mary if possible.

24 **DR. DANIEL SCHLENK:** Thanks. The panel  
25 recognizes Mary Mitchell.

26 **DR. MARY MITCHELL:** Good morning. I'm Mary



1 Mitchell. I'm Director General of the Environmental Director of  
2 Health Canada's Pest Management Regulatory Agency, which  
3 effectively makes me Don's counterpart in Canada.

4 I'd like to, first of all, thank the EPA for  
5 inviting Canada and California to be part of this event. Like  
6 Don, I'd like to acknowledge the collaborative efforts of the  
7 U.S. and the Canadian team. They really have worked well  
8 together to pool together the proposal you're going to be  
9 looking at.

10 I think this is an excellent example of good  
11 international collaboration. I strongly believe that  
12 international collaboration allows access to the best science  
13 available and ultimately results in more robust assessment  
14 methods and risk assessments.

15 The international work and pollinator  
16 collaboration is part of an ongoing series of collaborations  
17 that have been designed to improve and harmonize pesticide data  
18 requirements and assessment methods. It's harmonized data needs  
19 and assessment methods that make it possible for the U.S.,  
20 Canada and other countries to share the evaluation of scientific  
21 studies, which decreases regulatory burden for both regulated  
22 and regulated communities.

23 It also allows the coordinated and efficient  
24 pesticide registration decisions which results in better access,  
25 faster access to modern, safer pest control products for farmers  
26 in all the countries that are involved.



1           The U.S. and Canada have made it clear that  
2 we're highly committed to continuing such international  
3 collaboration, whether it be under the auspices of NAFTA, the  
4 OECD, or the more recent U.S., Canada Regulatory Cooperation  
5 Council that was jointly announced by President Obama and Prime  
6 Minister Harper last February.

7           And finally, I would like to thank you, the  
8 panel members for agreeing to participate. The breadth and  
9 depth of experience in this panel is really amazing. I'm not  
10 going to speak to the science; there will be plenty of other  
11 people doing that this week. The importance of ensuring  
12 pesticide use doesn't impact honey bee health goes without  
13 saying -- and I think agencies are really looking forward to  
14 your advice on how to further strengthen the proposed framework  
15 for characterizing potential effects of pesticides in bees. I'm  
16 quite sure this is going to be an interesting week. Thank you.

17           **DR. DANIEL SCHLENK:**       Thank you. Next on  
18 our agenda is Richard -- is it Bireley?

19           **MR. RICHARD BIRELEY:** Bireley, that is  
20 correct. My name is Richard Bireley. I work for the California  
21 Department of Pesticide Regulation as an ecotoxicologist. I,  
22 too, would like to thank the U.S. EPA for inviting staff from  
23 PRMA Canada and the California Department of Pesticide  
24 Regulation to participate in this Scientific Advisory Panel to  
25 address the potential risks of pesticides to honey bees.

26           The White Paper before you represents a



1 collaborative effort of OPP's Environmental Fate and Effects  
2 Division; Health Canada's Environmental Assessment Directorate  
3 and California DPR.

4 Just five years ago, following the review of  
5 data submitted as an adverse effects disclosure, staff at Cal  
6 DPR realized that the data requirements in 4D CFR were no longer  
7 adequate to address the uncertainties posed by pesticides to  
8 pollinators. It quickly became clear that this issue had been  
9 identified by staff at OPP's Environmental Fate and Effects  
10 Division and the Environmental Assessment Directorate at Health  
11 Canada.

12 As many of you are aware, California is the  
13 home of the largest pollination event in the world, when more  
14 than half the bee colonies in the United States are transported  
15 to California to pollinate over 800,000 acres of almonds.

16 I believe this international collaboration has  
17 resulted in a solid, focused approach that should also increase  
18 the confidence of the regulated community in a meaningful  
19 product that will meet international data requirements.

20 I would also like to thank each of you serving  
21 on the panel for agreeing to vet the proposal and provide input  
22 and strengthen it further. I know that Cal DPR staff have found  
23 this collaborative effort rewarding, and we all look forward to  
24 your input. Thank you.

25 **DR. DANIEL SCHLENK:** Thank you, Mr.  
26 Bireley. Okay. Our first presentation this morning will be



1 from Thomas Moriarty, who is in the Pesticide Reevaluation  
2 Division of OPP.

3 **MR. THOMAS MORIARTY:** Thank you, Chairman.  
4 And thank you, the FIFRA Scientific Advisory Panel, for allowing  
5 me to address you. My name is Tom Moriarty. I'm a risk manager  
6 in the Office of Pesticide Programs in the Pesticide  
7 Reevaluation Division.

8 During my presentation today, I will be  
9 providing an overview of pollinator declines and some of the  
10 efforts being conducted here in North American and abroad to  
11 refine exposure and effects assessment methods, as well as risk  
12 assessment methods for determining the potential impact of  
13 pesticides on pollinators and the role the pesticides play and  
14 pollinator declines in general.

15 I'll provide a brief overview of the current  
16 process used in North America to evaluate the potential effects  
17 of pesticides to bees and provide a brief overview of the risk  
18 assessment process itself, focusing on problem formulation phase  
19 where the protection goals are articulated.

20 Finally, I'll touch upon the extent to which  
21 honey bees serve as a surrogate for non-Apis bees and the extent  
22 to which a proposed risk assessment process is protective of  
23 non-Apis bees. This presentation relates to Charge Questions 1  
24 and 3 that deals with the protection goals in non-Apis bees.

25 A number of sources have reported declines in  
26 certain pollinator species globally. The 2006 report by the



1 National Academies of Science indicated decline in some North  
2 American pollinators, including North America's most important  
3 and productive managed pollinator, the honey bee.

4 However, at the time the NAS report was  
5 published, there was insufficient information to determine the  
6 cause of these declines. At roughly the same time, some of the  
7 declines were being reported in Europe in a report by Biesmeijer  
8 et al. in the July 2006 issue of Science.

9 The decline of managed honey bees has been  
10 measured by the U.S. Department of Agriculture's National  
11 Agricultural Statistics Survey. NASS data indicate that managed  
12 honey bee colonies have declined from a peak of approximately 6  
13 million colonies in 1947 to roughly 2.8 million colonies in  
14 2006.

15 These declines reflect a variety of factors in  
16 North American society and agriculture, as well as changes in  
17 the way that a NASS itself collect survey data. Despite changes  
18 in any survey techniques, data indicates that the overall number  
19 of managed colonies had steadily declined over the past 60  
20 years.

21 Although not depicted on the graph here, the  
22 most recent estimate of 2011 places the number of colonies in  
23 the U.S. at roughly 2.5 million. One of the factors affecting  
24 honey bee declines has been introductions, spread of Tracheal  
25 and Varroa mites. Prior to the introduction of mites, average  
26 colony of losses were around five to ten percent; however, since



1 the introduction of parasitic mites in the U.S. in the 1980s,  
2 typical losses of managed honey bee colonies have roughly been  
3 about 15 to 21 percent.

4 According to USDA, starting around 2007,  
5 average colony loss suddenly increased and ranged between 31 and  
6 36 percent. Losses varied among beekeepers, and for some,  
7 losses were complete.

8 Some of these losses were marked by sudden  
9 loss of adult forage bees, leaving only the queen behind, a few  
10 nurse bees, developing brood and ample food reserves. This  
11 scenario would lead to the eventual collapse of the colony and  
12 was termed Colony Collapse Disorder.

13 While large losses in 2006 and 2007 were  
14 attributed to CCD, per se, the contribution of CCD to overall  
15 declines in managed colonies appears to be on the decline.  
16 Reports of CCD symptoms continue, however, declines in honey bee  
17 health are attributed to symptoms such as winterkill, failure to  
18 thrive and queen loss.

19 While a number of factors and agents have been  
20 hypothesized as potential contributors to CCD and general  
21 pollinator declines, at this time, no factor has been identified  
22 as single cause. Available science suggests that pollinator  
23 declines are a result of multiple factors which may be acting in  
24 various combinations.

25 USDA, the lead federal agency on investigating  
26 general declines in pollinator health has hypothesized that



1 losses of managed colonies may be caused by primary stressors  
2 such as parasitic mites, poor bee management, nutrition and/or  
3 pesticides that may, in turn, cause honey bees to become  
4 susceptible to disease and ultimately lead to the collapse of  
5 the colony.

6 Research continues to be directed at the range  
7 of individual factors, but is also being directed at identifying  
8 combinations of stressors that are most strongly associated with  
9 pollinator declines. As part of the federal response, a general  
10 decline in pollinators, EPA's Office of Pesticide Programs has  
11 taken a number of steps to improve the understanding of the role  
12 of pesticides and pollinator declines. As part of those  
13 efforts, EPA, along with its state and federal partners, has  
14 developed a proposed pesticide risk assessment framework for  
15 pollinators.

16 Over the past several years, EPA has actually  
17 been actively engaged in a number of efforts, both here and  
18 abroad, aimed at improving and refining the methods for  
19 examining the potential adverse effects of pesticides to bees.

20 In 2009, EPA hosted a USDA-funded meeting to  
21 discuss testing protocols for acute and chronic toxicity studies  
22 with honey bees. Participants identified uncertainties and  
23 limitations of existing protocols and considered refinements to  
24 those test protocols. Participants also discussed moving test  
25 designs toward more comprehensive and standardized protocols to  
26 increase consistency and reproducibility.



1 In 2011, the Society of Environmental  
2 Toxicology and Chemistry, abbreviated as SETAC, sponsored a  
3 global Pellston Workshop on pesticide risk assessment for  
4 pollinators. The intent was to synthesize the best available  
5 science regarding pesticide exposure and effects assessment  
6 methods for Apis and non-Apis, and to further harmonize risk  
7 assessment approaches among regulatory authorities.

8 The Pellston Workshop had four major themes:

- 9 1. Exposure Assessment.
- 10 2. Laboratory Effects Assessment
- 11 3. Field Effects Assessment
- 12 4. Integrating Exposure in Effects Data to  
13 estimate the likelihood and magnitude of  
14 potential effects known as risk assessment.

15 A fifth area of focus was on determining the  
16 extent to which honey bee exposure and effects estimates could  
17 serve as a reasonable measures with which to evaluate risk to  
18 non-Apis bees. The International Commission for Plant Bee  
19 Relationships, abbreviated at ICP-BR, which is affiliated with  
20 the International Union of Biological Science, draws upon  
21 expertise and government industry and academia to inform  
22 scientific assessment aimed at bee protection. ICP-BR has been  
23 developing methods for testing the toxicity of pesticides to  
24 bees and these efforts have informed testing methods supported  
25 by the European and Mediterranean Organization for Plant  
26 Protection, abbreviated as EPPO, and the Organization for



1 Economic Cooperation and Development, OECD.

2           The prevention of colony losses, abbreviated  
3 as COLOSS, is a global network of scientists, beekeepers and  
4 industry members from 55 countries, focused on developing  
5 standards for monitoring and research on factors associated with  
6 colony losses.

7           The OECD's working group on pesticides has  
8 also developed an expert subgroup on pollinators, entitled PEIP,  
9 Pesticide Effects on Insect Pollinators workgroup. EPA and PMRA  
10 serve as co-chairs to the PEIP workgroup and have been working  
11 with other member countries on issues related to pollinator  
12 protection. The focus of the PEIP workgroup developed from a  
13 2010 OECD member survey on regulatory needs related to  
14 pollinators that identified four themes. These themes included  
15 advancing and harmonizing the science of risk assessment,  
16 developing a source for information on risk management  
17 approaches, developing tools to share bee kill incident  
18 information, and providing an index of research relevant to  
19 pollinator health and protection.

20           Finally, in 2012, the European Food Safety  
21 Authority's panel on plant protection products and their  
22 residues release a scientific opinion on the development of risk  
23 assessment processes for bees, including Apis as well as  
24 non-Apis.

25           The opinion examined the potential role of  
26 exposure, strengths and limitations of current toxicity testing



1 methods and provided recommendations on how to improve these  
2 methods. Currently, OPP relies on a qualitative process for  
3 evaluating the potential hazard of pesticide to honey bees.

4 Typically, the quantified risks to non-target  
5 organisms, the Agency relies on point estimates of exposure and  
6 effects and the ratio of those estimates, referred to as the  
7 risk quotient. Because not every organism can be tested,  
8 toxicity testing is typically conducted using surrogate species.

9  
10 Selection of a surrogate species is based on  
11 the extent to which they are readily available and amenable to  
12 testing under laboratory and field conditions. This then  
13 reflects the species for which husbandry conditions are well  
14 defined. For evaluating potential hazard to terrestrial  
15 invertebrates, insect pollinators, toxicity tests are frequently  
16 conducted, used in Western or European honey bee.

17 The test listed on this slide will be  
18 discussed in greater detail in the presentation to follow. I'll  
19 only make several notes on this current tests used in North  
20 America to estimate hazard to honey bees. The toxicity tests  
21 are typically tiered, where the first tier is an acute toxicity  
22 test with young adult bees.

23 According to EPA test requirements, if the  
24 acute contact toxicity is less than 11 micrograms per bee and  
25 toxicity of residues on foliage may be required. If the  
26 toxicity is less than 11 micrograms per bee and there is



1 evidence of prolonged exposure and/or if there is evidence that  
2 bee colonies may be adversely affected, then a field study may  
3 be required.

4 Unlike PMRA, EPA guidelines do not currently  
5 address acute oral toxicity to capture potential exposure  
6 through residues in pollen or nectar that may result from  
7 systemic pesticides; nor do current guidelines address the  
8 potential toxicity of pesticides to larval bees. However, acute  
9 oral toxicity tests are available in the OECD Guidelines, and  
10 while acute toxicity tests with larval bees are continuing to  
11 evolve, several methods are also currently available.

12 As it will become apparent through the  
13 presentations to follow, the closed framework makes use of all  
14 the current guideline studies and recommends the addition of  
15 certain tests to address areas of uncertainty.

16 The figure above represents the overall risk  
17 assessment process. This process is consistent with EPA  
18 guidelines and is consistent with a process used by our  
19 partners, PMRA and California DPR. It consists of three phases  
20 that include problem formulation, analysis, which consists of  
21 exposure and effects assessment, and risk characterizations,  
22 consisting of risk estimation and risk description.

23 Throughout the assessment process, multiple  
24 lines of evidence are synthesized, and although the figure  
25 suggests that the process is unidirectional, it is intended to  
26 be iterative where exposure and toxicity data reviewed in the



1 analysis phase may be used to refine the risk hypothesis  
2 articulated in the problem formulation phase, or where risk  
3 estimates derived in the risk characterization phase may be  
4 refined through additional data from the analysis phase.

5 For the remainder of the presentations,  
6 detailed elements of this process will be discussed. However,  
7 before doing so, I would like to draw your attention to the box  
8 at the top left of the diagram, entitled, "Planning." As it  
9 indicated, the risk assessment process is intended to be  
10 iterative and intended to involve both risk assessor and risk  
11 managers.

12 The planning of the risk assessment is  
13 intended to occur early in the process as an opportunity for  
14 risk managers and risk assessors to articulate management goals  
15 and whether there are complexities or issues that may require  
16 particular attention. These discussions, in turn, inform  
17 decisions regarding resources and scheduling needed to complete  
18 the assessment.

19 The presentations today will highlight the  
20 tiered nature of the risk assessment process. The initial tier  
21 serves as a screen where conservative assumptions about exposure  
22 and effects are made, and for those chemicals which do not  
23 indicate a minimal risk at the screen level, additional analysis  
24 in the form of refinements or higher tiers may be necessary.

25 While the decision to proceed to higher levels  
26 of refinement depend on the understanding of both the risk



1     assessor and risk manager with regard to the management needs.  
2     Frequent discussions between the risk assessor and the risk  
3     manager often yields opportunities where data or information  
4     needs are identified early in the process and it can be  
5     communicated to the technical registrant or the appropriate  
6     stakeholder.

7             At the top of the figure, the problem  
8     formulation phase is identified. The formula is intended to  
9     identify the objective of the risk assessment and produce three  
10    products.

11            The assessment and measurement endpoints that  
12    reflect management goals, conceptual models that describe key  
13    relationships between the stressor and the assessment endpoint  
14    and the analysis plan. Other presentations you'll hear today  
15    will discuss a conceptual model and analysis plan aspects of the  
16    problem formulation. But over the next several slides, I'll  
17    discuss goals and endpoints and how they are used in the process  
18    and their relationship to one another.

19            Ecological risk assessment is typically  
20    developed in a risk management context to evaluate human-induced  
21    changes that are considered undesirable. Changes often  
22    considered undesirable are those that alter important structural  
23    or functional characteristics or components of an ecosystem. An  
24    evaluation of adverse affects may include consideration of type,  
25    intensity and scale of the effects and well as the potential for  
26    recovery.



1                   Acceptability of certain adverse effects  
2 reflects risk management. It is done so in the light of multiple  
3 factors used to weigh both the risks and the benefits of a  
4 pesticide. Therefore, it's important that as a risk assessment  
5 is being developed, both management goals and science objectors  
6 are considered and specificity is given when developing  
7 assessment endpoints in conceptual models.

8                   Protection goals are statements by a  
9 regulatory authority on the desired condition of values of  
10 concern. When they are stated at the highest levels, they may  
11 be considered generic. U.S. EPA's generic protection goal is to  
12 protect human health and the environment. With respect to  
13 pesticides, management goals are in part defined by the laws  
14 that would provide statutory authority to regulate them.

15                  The Federal Insecticide Fungicide and  
16 Rodenticide Act directs EPA to regulate pesticides, such that  
17 their use does not result in unreasonable adverse effects on man  
18 or the environment, taking into account economic, social an  
19 environmental costs and benefits from the use of any pesticide.  
20 These generic protection goals therefore reflect statutory  
21 scientific and societal goals of a regulatory agency.

22                  Generic protection goals, however, do not --  
23 may not provide adequate guidance at the risk assessment level.  
24 And specific protection goals may be needed which can better  
25 direct and inform decisions at that level. Specific protection  
26 goals speak more directly to the objectives for an ecological



1 entity and inform the type of data that is needed; so the  
2 specific protection goals therefore must be consistent and  
3 support generic protection goals.

4 Consistence with the results of the SETAC  
5 Pellston Workshop of specific protection goals with this  
6 proposed risk assessment framework include protection of  
7 pollination services provided by bees; protection of honey  
8 production and other hive products, and the protection of  
9 pollinator diversity, in terms of adequate number and diversity  
10 species that contribute to the health of the environment.

11 In its recent scientific opinion on the  
12 development of risk assessment of plant protection products for  
13 bees, the European Food Safety Authority has also identified  
14 pollination, hive products and biodiversity as relevant  
15 ecosystem services and values to protect.

16 Once protection goals are identified, specific  
17 assessment and measurement endpoints can be developed that are  
18 consistent with and support the stated protection goals. As  
19 defined in the EPA guidelines, assessment endpoints are explicit  
20 expressions of the actual environmental values that are to be  
21 protected and are operationally defined as an ecological entity  
22 and its attributes.

23 The ability of assessment endpoints to support  
24 risk management decisions is dictated by whether or not there  
25 are measurable characteristics that adequately represent or  
26 reflect management goals. EPA has identified three criteria for



1 determining the selection of assessment endpoints and it  
2 included whether they are ecologically relevant, susceptible to  
3 the known or potential stressor, and what's relevant to the  
4 management goals.

5 Assessment endpoints are then specifically  
6 informed by the data generated for the risk assessment. Both  
7 effects data and exposure data provides specific information as  
8 measurement endpoints than inform one or more of the assessment  
9 endpoints.

10 When measurement endpoints are appropriately  
11 linked to assessment endpoints and specific protection goals,  
12 they therefore also support generic protection goals. This  
13 slide is Table 1 of the White Paper and illustrates both  
14 examples of and the relationship between measurement endpoints,  
15 assessment endpoints and specific protection goals.

16 The specific protection goals identified in  
17 the table are then consistent with the generic protection goals  
18 of protecting human health and the environment. The measurement  
19 endpoints identified in the table are not intended to be  
20 exhaustive, but only serve as examples. For the remaining  
21 presentations, a range of measurement endpoints will be  
22 discussed in the context of the pro's risk assessment framework.

23  
24 In conclusion, the presentations you will hear  
25 today will layout a proposed risk assessment framework that is  
26 tiered, iterative and flexible, allowing for multiple lines of



1 evidence to be considered and refinements to be made. In this  
2 way, the proposed framework is sensitive with respect to the  
3 resources needed to complete a risk assessment.

4 The primary focus of the proposed framework is  
5 for evaluating the potential for adverse effects to honey bees  
6 and also uses a honey bee as a surrogate for other non-Apis  
7 bees. The extent to which any species is a reasonable surrogate  
8 for other species, involves considerable uncertainty, unless  
9 sufficient data are available to put differences in  
10 relationships into context.

11 Uncertainties reflect differences in life  
12 biologies and histories that can impact both exposure and  
13 effects. Despite the potential differences among bee species,  
14 it is believed that the proposed risk assessment process with  
15 honey bees provides information on non-Apis bees as well. At  
16 lower tiers, for example, where effects to individual bees is  
17 tested, exposure routes considered, i.e. contact and ingestion  
18 are shared by both honey bees and non-Apis bees alike.

19 Until guideline studies with non-Apis bee  
20 species are better developed and available for regulatory  
21 purposes, the Agency is asking the panel the extent to which it  
22 believes the proposed processed is informative of a potential  
23 pesticide risk to non-Apis species.

24 Finally, EPA and its partners seek the opinion  
25 of the panel regarding the extent to which the proposed risk  
26 assessment framework supports the identified protection goals



1 and the extent to which biodiversity, identified as a protection  
2 goal, is supported by the use of a honey bee, insofar as it is a  
3 reasonable surrogate for non-Apis bees.

4 Thank you for your time. And if there are no  
5 questions, I would like to allow Mr. Sappington to move into his  
6 presentation on conceptual models.

7 **DR. DANIEL SCHLENK:** I think we do have some  
8 questions. Again, this is a general overview. So if you could  
9 hold your questions to more of the general framework, I believe  
10 future speakers will have very specific details on the process.  
11 So if you have questions related to the overall objectives,  
12 then, yeah. Dr. James.

13 **DR. ROSALIND JAMES:** I have a simple question.  
14 You talked about management goals. The word "management" has a  
15 lot of meanings to me. Are you talking about pest management  
16 goals in your model framework?

17 **MR. THOMAS MORIARTY:** Risk management goals,  
18 balancing between the risks and benefits.

19 **DR. DANIEL SCHLENK:** That was Mr. Moriarty.  
20 Let me just remind the speakers that anytime you respond, can  
21 you state your name real quick beforehand so that we can get  
22 that on the written record. Thank you.

23 Any other questions?

24 (No response.)

25 Okay. Mr. Sappington, do you want to take the  
26 reins for the next presentation? We can get that loaded.



1                   **MR. KEITH SAPPINGTON:** Hello. My name is  
2 Keith Sappington. I'm a biologist and senior science advisor in  
3 the ecological Environmental Fate and Effects Division within  
4 the Office of Pesticide Programs.

5                   I appreciate this opportunity to address the  
6 FIFRA Scientific Advisory Panel on this important topic, and I  
7 look forward to your feedback to help us improve our process for  
8 pesticide risk assessment to bees.

9                   This morning I'll be summarizing two aspects  
10 of the White Paper in back-to-back presentations. In the first  
11 presentation, I will describe a set of generic conceptual models  
12 we are proposing for assessing pesticide risk to honey bees.

13                  Details of these generic conceptual models are  
14 found in Section II of the White Paper. Immediately following  
15 this presentation I will summarize our proposed decision  
16 framework for assessing risks of pesticides to honey bees. And  
17 this is also found in Section II of the White Paper.

18                  With respect to conceptual models, I will  
19 first provide a brief overview of the use of conceptual models  
20 and ecological risk assessment, and then I'll describe the  
21 context and the purpose of these models as we apply them in  
22 ecological risk assessment of pesticides to honey bees.

23                  Following this introduction, I'll then  
24 describe the generic conceptual models that are specific to four  
25 common application methods. And these include foliar spray  
26 applications for both systemic and non-systemic pesticides, soil



1 application for systemics, and seed treatment application for  
2 systemic pesticides.

3 We present a fifth conceptual model in the  
4 White Paper for trunk drench and tree injection, but I will not  
5 be presenting that this morning.

6 The figure on the right of this slide  
7 illustrates the components of problem formulation phase and it  
8 depicts, visually, how the selection of assessment endpoints and  
9 conceptual models are used to inform the analysis plan of the  
10 risk assessment.

11 As defined in EPA's ecological risk assessment  
12 guidelines, a conceptual model is a written and visual  
13 description of the hypothesized relationships between the  
14 ecological entities and the stressors of concern. In the  
15 conceptual models I'll be presenting today, the ecological  
16 entities, or honey bees, and the stressor of concern is the  
17 pesticide application, and this includes any degradates that are  
18 of toxicological concern.

19 The written portion of a conceptual model  
20 includes a set of risk hypotheses which are statements regarding  
21 the purported relationships between the entities and the  
22 stressor. The visual portion of the conceptual model -- which I  
23 will be presenting shortly -- are diagrams of these purported  
24 relationships.

25 In the context of ecological risk assessment,  
26 conceptual models are useful tools for communicating to risk



1 managers, risk assessors and the public of how we believe  
2 exposure to pesticides and their associated effects are likely  
3 to occur.

4 With that said, there is not a single standard  
5 format for depicting a conceptual model diagram. They can vary  
6 widely in their level of detail, from simple, highly aggregated  
7 diagrams to ones that are very complex and highly disaggregated.

8  
9 In addition, the graphical format can vary,  
10 for example, from the more traditional box and arrow diagrams,  
11 to ones that are pictorial in their representation. But  
12 regardless of format, the general structure of conceptual models  
13 is generally similar and, as indicated on the bottom of this  
14 slide, represents a cascade of events which begins at the  
15 stressor source and its relevant exposure routes, and then to  
16 receptors of concern and their responses, and ultimately to  
17 changes in the attributes or the assessment endpoints  
18 identified in the risk assessment.

19 The generic conceptual models I'll be  
20 presenting this morning are consistent with the format typically  
21 used in EPA pesticide risk assessments. Before presenting the  
22 conceptual models, I first would like to make several points  
23 regarding their development and intended use in risk assessment.

24  
25 First, the generic conceptual models are  
26 called generic because they're intended to be used as a starting



1 point for pesticide risk assessors. And therefore, are expected  
2 to be modified, where necessary, depending on the circumstances  
3 of that specific risk assessment.

4 Second, the conceptual models presented here  
5 are not comprehensive of all pesticide uses. Rather, they  
6 represent several major use categories that are expected to  
7 result in some exposure to honey bees.

8 Lastly, in addition to being organized around  
9 different pesticide use patterns, the models also consider some  
10 differences in chemical characteristics, and in particular, the  
11 systemicity of a pesticide, which, in turn, impacts the nature  
12 of exposure to honey bees.

13 In describing the generic conceptual models, I  
14 first plan to go through in detail the conceptual model for  
15 foliar applications of non-systemic pesticides here and then  
16 I'll highlight the main differences for the other three  
17 conceptual models that we have identified.

18 As shown in bold, on the left, these  
19 conceptual models begin at the top, with the stressor of concern  
20 and continue downward by illustrating the sources of exposure,  
21 the related exposure media, receptors of concern and changes in  
22 the attribute or assessment endpoints.

23 In this case, the stressor of concern is a  
24 non-systemic pesticide application -- and again, including any  
25 of its degradates -- that is applied to a crop foliage, be it  
26 ground or application methods. As shown in these boxes, the



1 exposure sources considered in this model include deposition of  
2 spray droplets directly onto bees, plants, soil, and surface  
3 water, either on the treated site or via drift, to adjacent  
4 sites.

5 From these exposure sources, resulting  
6 pesticide residues in various environmental media are  
7 considered, including residues on plant surfaces, in soil,  
8 pollen and nectar, and in surface water, which can result from  
9 spray drift and pesticide runoff/erosion, shown in blue.

10 You'll notice that the dashed line surrounding  
11 the residues in the soil and surface water boxes. This  
12 indicates that we believe these exposure pathways are less  
13 important to honey bees compared to other exposure routes  
14 depicted. Additional information supporting this distinction  
15 will be presented later in a presentation by Ms. Christina  
16 Wendel.

17 The nature of pesticide exposure is then  
18 depicted by the lines connecting the pesticide residues and  
19 environmental media to the receptors. Worker bees foraging for  
20 food and water sources constitute the primary receptor of  
21 concern outside of the hive; although direct exposure to drones  
22 and the queen is also possible during mating and orientation  
23 flights.

24 Primary routes of exposure outside of the hive  
25 that we have included include dermal uptake of pesticide, via  
26 spray droplets and foliar residues, inhalation -- which is



1 applicable to more volatile pesticides, primarily -- ingestion  
2 of pesticides absorbed onto pollen and nectar.

3 Secondary routes of exposure of lessor concern  
4 for honey bees include contact with contaminated soil and  
5 surface water. Another secondary route of exposure not depicted  
6 here is contaminated dew on plant surfaces. Exposure from  
7 contaminated dew droplets is discussed in detail in a later  
8 presentation.

9 We note, however, that exposure from pesticide  
10 residues in soil is likely to be more important for some  
11 non-Apis bee species. For example, ground nesting bees that  
12 collect and process soil for nest construction.

13 From here, exposure to bees inside the hive is  
14 possible, via direct contact, ingestion and processing of  
15 contaminated pollen and nectar brought into the hive by the  
16 workers. Ingestion of honey and as well as processing of wax  
17 and propolis during comb production and brood rearing activities.

18  
19 Exposure of hive bees to pesticide is also  
20 possible from contaminated water used for consumption, honey  
21 production, and evaporative cooling. And this pathway is  
22 further described in a later presentation.

23 Subsequently, bee brood may be exposed to  
24 pesticides via ingestion of brood provisions that includes brood  
25 food, honey, and processed pollen, wax and propolis. Inside the  
26 hive, queens are expected to be exposed primarily through



1 contact with comb material and consumption of royal jelly.

2 Lastly, the relationships between the exposure  
3 of different bee castes and changes in attributes or assessment  
4 endpoints are shown with these arrows and text boxes. The text  
5 in bold in these boxes represent either the assessment endpoints  
6 or in a case to pollinator biodiversity, the protection goal.  
7 Example of measurement endpoints are shown below at each of the  
8 assessment endpoints.

9 We note that these arrows collapse many  
10 complex chemical, biological and toxicological processes that  
11 collectively determine the responses of individual bees and the  
12 entire colony to a given pesticide exposure. For example, the  
13 populations size and stability of colonies may not only be  
14 impacted by a direct loss in forager bees due to mortality, but  
15 also by other factors, including, but not limited to reduced  
16 foraging success and associated reduction in food reserves,  
17 insufficient workers to maintain hive temperatures during  
18 winter, and a reduction in homing success.

19 For systemic foliar applied pesticides, the  
20 generic conceptual model is identical to that described  
21 previously for non-systemic pesticides, except that it includes  
22 pesticide uptake and translocation to the plant tissues. And I  
23 show this in red in the arrows in this figure.

24 Thus, for systemic pesticides that are applied  
25 to foliage via spray method, a portion of the residues in pollen  
26 and nectar may result from pesticide deposition onto these



1 tissues in addition to a portion that may be subsequently  
2 transported to these tissues via translocation.

3 The remainder of this conceptual model from  
4 the receptors downward is the same as described previously for  
5 non-systemic foliarly applied pesticides. For application of  
6 systemic pesticides directly to bare soil either by granular  
7 spray or soil incorporation methods, the dominate route of  
8 exposure we have depicted for honey bees begins with pesticides  
9 uptake and translocation within the developing plant.

10 As with the foliarly-applied systemic  
11 pesticides, the foraging bees may then be exposed from ingestion  
12 and contact with residues in pollen, nectar, plant exudates, and  
13 honey dew. It is assumed that direct exposure of foraging bees  
14 with pesticide spray droplets would not be a primary exposure  
15 route of concern because soil applications would be presumed to  
16 occur prior to planting.

17 Furthermore, spray applications to bare soil  
18 typically involve a coarse droplet spectrum which would not  
19 likely to be subject to extensive offsite drift. However, if  
20 offsite drift of spray droplets was a concern, or if there was  
21 attractive foliage in the field at the time of the soil  
22 application, then these exposure routes would be addressed as  
23 described previously for the foliar spray conceptual model.

24 Runoff of a pesticide into surface water is  
25 another potential route of concern, but this is expected to be  
26 minor relative to exposure from residues in pollen and nectar.



1 Again, the remainder of the conceptual model  
2 from the receptors downward is the same as described previously.

3 Exposure of honey bees to pesticides via seed treatment is  
4 depicted here as a result of two major sources: offsite drift of  
5 abraded seed coat dust and from pesticide residues in plants  
6 that result from pesticide translocation to pollen, nectar,  
7 plant exudates and honeydew.

8 Subsequent exposure of bees to pesticides in  
9 these matrices follows the same pathways as depicted earlier for  
10 soil application of systemic pesticides. For drift of abraded  
11 seed coat dust, foraging bees may be exposed by directly  
12 intercepting dust particles containing the pesticide during  
13 flight and subsequent dermal or respiratory uptake. It is also  
14 possible for contaminated dust to deposit on other environmental  
15 media, including soils, surface water, and plant tissues.

16 Generally speaking, these routes are likely to  
17 result in lower exposures compared to direct contact with seed  
18 dust or contaminated pollen and nectar from the treated crop.

19 We note, however, that exposure to pesticides  
20 from deposition of contaminated dust particles onto flowing  
21 plants may be important, particularly if these plants are  
22 located in close proximity to the seed planting operations.  
23 Again, the bottom half is the same as I described earlier.

24 So to conclude, the generic conceptual models  
25 for pesticide exposure and effects on honey bees presented here  
26 and in the White Paper, are intended to reflect only several



1 major pesticide use categories. They also are intended to serve  
2 as a basis for consideration in a forthcoming pesticide risk  
3 assessment involving honey bees, and are expected to be modified  
4 when circumstances warrant.

5 As indicated in Charge Question Number 2, we  
6 are seeking feedback on these models, in terms of their overall  
7 accuracy and the extent to which their implementation is  
8 consistent with the proposed assessment endpoints and management  
9 goals.

10 At this time, myself and the team would  
11 address any clarifying question on the generic conceptual  
12 models. And after that, I will present on our proposed decision  
13 frameworks. Thank you very much.

14 **DR. DANIEL SCHLENK:** Thank you. Okay, any  
15 questions of clarifications. Dr. Potter?

16 **DR. THOMAS POTTER:** I have a question about  
17 your category of foliar-applied pesticide. Does that  
18 essentially include all non-systemic broadcast applied active  
19 ingredients?

20 **MR. KEITH SAPPINGTON:** For the first  
21 conceptual model I presented, that's for non-systemic  
22 foliar-applied sprays.

23 **DR. THOMAS POTTER:** But does it have to be  
24 foliar applied is, I guess, where I'm looking for an answer on  
25 that?

26 **MR. KEITH SAPPINGTON:** Well, we distinguish



1 between foliar and soil applied. So the presumption is that it  
2 is being applied to foliage in that conceptual model.

3 **DR. THOMAS POTTER:** But it seems to leave out  
4 a large category of active ingredients that one presumes  
5 assessments being made regarding risks, pre-emergent herbicides  
6 would be one category.

7 **MR. KEITH SAPPINGTON:** Right. Those would, I  
8 think, be covered in the soil application methods; the  
9 conceptual model for soil application.

10 **DR. THOMAS POTTER:** I guess what I saw for  
11 soil application, though, was systemics and not, you know, non -  
12 again, non-systemics are what we're talking about here. Again,  
13 I'm just looking for clarification on this as to where those  
14 categories of chemicals that are soil applied, that are  
15 non-systemic, where do they fall into these four different  
16 categories?

17 **MR. KEITH SAPPINGTON:** For non-systemic  
18 soil-applied pesticides, you would be primarily concerned about  
19 the contact exposure; although, again, if they're applied  
20 pre-emergence to bare soil then the exposure to bees foraging in  
21 that particular field site would be presumed to be rather low.  
22 If there were foliage present then it would follow the foliar  
23 conceptual model. Or, if there was a reason to expect drifts  
24 offsite from that, then it would follow the foliar conceptual  
25 model framework.

26 **DR. THOMAS POTTER:** So again, just to be



1 clear, all non-systemic active ingredients, which would be  
2 broadcasts applied, would fall into that first schematic that  
3 you showed?

4 **MR. KEITH SAPPINGTON:** I think that would be  
5 the closest one that would cover them. Now we could split those  
6 out as we refine these conceptual models to make that more  
7 clear. But we focused on the systemics because of the focus on  
8 the uptake in translocation in the plants and exposure through  
9 pollen and nectar. But we can break that out to make that  
10 clearer. Thank you.

11 **DR. DANIEL SCHLENK:** Any others? Yes, Dr.  
12 McManaman.

13 **DR. JAMES MCMANAMAN:** I see that larvae left  
14 out of your model here, it doesn't look like they're present in  
15 there. Is that to simply the model or is that because you have  
16 in mind a different kind of model for larvae?

17 **MR. KEITH SAPPINGTON:** The larvae would be  
18 represented towards the bottom of the conceptual model as part  
19 of the brood in the hive. That's where we would be expecting  
20 their exposure for honey bees.

21 **DR. JAMES MCMANAMAN:** So you make a  
22 distinction between the brood provisions and provisions for the  
23 queen, in terms of royal jelly. But my understanding is that  
24 oftentimes early on, the bees receive the same kind of food  
25 until it's distinguished between who's going to become queen or  
26 not. Is that again left out due to simplification?



1                   **MR. KEITH SAPPINGTON:** Let me see if I can  
2 understand the question first. So the question is, is direct  
3 exposure from pollen and nectar to bee brood as opposed to its  
4 processed pollen and nectar and how is that represented in the  
5 conceptual model. Do I understand it correctly?

6                   **DR. JAMES MCMANAMAN:** Yes.

7                   **MR. KEITH SAPPINGTON:** Yes. My understanding  
8 is that was left out just for simplicity reasons. I mean, at  
9 some point you get to have so many hours and lines directed. My  
10 understanding is that -- and we've investigated and I believe  
11 Ms. Kris Garber will be presenting more information about the  
12 consumption of larvae later today -- but we've investigated the  
13 extent to which the larvae were consuming or may consume pollen  
14 directly versus the process pollen.

15                   **DR. JAMES MCMANAMAN:** So the Agency's view is  
16 that larvae are included in this model system and the details  
17 may --

18                   **MR. KEITH SAPPINGTON:** Absolutely.

19                   **DR. JAMES MCMANAMAN:** -- vary from adults.

20                   **MR. KEITH SAPPINGTON:** Absolutely.

21                   **MR. JAMES MCMANAMAN:** Okay.

22                   **DR. DANIEL SCHLENK:** Any other questions.  
23 Clarification? Dr. James?

24                   **DR. ROSALIND JAMES:** This is Rosalind James.  
25 I think this is the appropriate place to ask this question.  
26 When honey is made, nectar is collected and then a significant



1 proportion of the water is evaporated off and it becomes  
2 concentrated. Is that included in your analysis at all?

3 **MR. KEITH SAPPINGTON:** We have a separate  
4 presentation this afternoon that looks at the whole issue of  
5 contaminated surface water and its relative importance, compared  
6 to these other aspects.

7 **MS. ROSALIND JAMES:** I'm actually talking  
8 about the nectar itself. So the honey is made from nectar and  
9 then the nectar is predominately water. But it's the nectar --  
10 I'm not talking about collected water. And then to make honey,  
11 the bees evaporate off the --

12 **MR. KEITH SAPPINGTON:** Right.

13 **MS. ROSALIND JAMES:** I mean, that's processing  
14 of the nectar. But is the fact that you're concentrating the  
15 nectar down included in your risk assessment? It could change  
16 the exposure level.

17 **MR. KEITH SAPPINGTON:** That's right. There  
18 are basically two competing processes, potentially, as we  
19 understand them. One would be degradation of the pesticide over  
20 time. And then conceptually, there could be some concentration  
21 as the honey is formed. I personally am not aware of data on  
22 that concentration phase, but those processes would be, I think,  
23 in competition with one another.

24 **DR. THOMAS STEEGER:** I would like to address  
25 Dr. James's question. As you will learn from the presentation  
26 from Kris Garber later on today, the concentration of residues



1 that could potentially take place as honey is being processed  
2 into the colony is accounted for in our exposure modeling  
3 because it's expressed on a sugar basis, as opposed to a water  
4 basis. So hopefully after you hear that presentation you'll  
5 have a better understanding of how we've accounted for that  
6 potential mechanism of exposure.

7 **DR. DANIEL SCHLENK:** Thank you, Dr. Steeger.  
8 Any other questions? Dr. Fefferman.

9 **DR. NINA FEFFERMAN:** My guess is that that the  
10 answer to this will be addressed in a later portion. But I was  
11 wondering, in the conceptual modeling where you felt that the  
12 layer for how the impact might propagate throughout a colony, it  
13 looks to me like everything that you've discussed is sort of  
14 exposure to individual bees and how that might get to individual  
15 bees. Where do you see that layer fitting?

16 **MR. KEITH SAPPINGTON:** I agree. As I went  
17 through the conceptual model I noted that those lines would  
18 connect the exposure media to the receptors and ultimately to  
19 the changes in the attributes, collapse a lot of information  
20 down. One aspect that I've thought out, personally, you know,  
21 there's no reason that a conceptual model necessarily has to fit  
22 on one page.

23 So we could have a conceptual model, much like  
24 this, that addresses the exposure pathways and then a separate  
25 one that lays out the various ways in which the response is to  
26 that given exposure, could then have affects on the hive. And



1 that could be very useful for the planning phase of a risk  
2 assessment. Thank you for that comment.

3 **DR. NINA FEFERMAN:** Is that currently in some  
4 section somewhere for consideration or is that something that  
5 really might be flushed out better in the conceptual model  
6 section but separately, as you proposed?

7 **MR. KEITH SAPPINGTON:** We discuss, texturally,  
8 in the White Paper these various ways in which the colony  
9 responses could be manifested, but they're not visually  
10 presented, currently.

11 **DR. THOMAS STEEGER:** I would like to add to  
12 Mr. Sappington's response by saying that as you will learn in a  
13 later presentation, it's not captured in the conceptual model,  
14 but we evaluate multiple lines of evidence to estimate, not just  
15 risk to individual bees, but to the colony as a whole. And part  
16 of that process we will be asking for SAP input on, is the use  
17 of colony level modeling in addition to the advanced or more  
18 refined testing that is conducted at the semi-field and full  
19 field level that look at whole colony effects.

20 **DR. DANIEL SCHLENK:** Okay. Any other  
21 questions before we break?

22 (No response.)

23 All right. Thanks. We're going to go by this  
24 clock on the wall here. Let's be back at 10:35.

25 (Brief recess.)

26 **DR. DANIEL SCHLENK:** Okay. Thanks. Again,



1 let me just reiterate, if you're on the phone, please mute your  
2 line at this time so we don't hear you during the presentations.

3  
4 Before we go to our next presentations, I  
5 believe there were some questions, some overall framework  
6 questions that were not asked the last time, for you, Keith, if  
7 that's okay. I believe Dr. Fefferman has questions, please.

8 **DR. NINA FEFFERMAN:** So forgive me, these  
9 don't actually address the specific conceptual models you were  
10 putting forth because I think those are really quite complete  
11 and lovely, and this is more a structural, as we consider the  
12 framework of risk assessment itself type of question.

13 In the types of risk assessment frameworks  
14 that I'm used to considering, sort of the very first step is  
15 which things are going to be useful to measure and which things  
16 are going to inform choices and how do we balance those. And I  
17 feel sort of as though we skipped past that to here are the  
18 things we're definitely measuring and here are the conceptual  
19 models and how those measurement might play out in terms of the  
20 risks associated with those particular aspects.

21 Has there been a stage of evaluation about  
22 some of these? I mean, I know we're not discussing necessarily  
23 the benefits of some of the pesticide use, but some of the risks  
24 to colonies, to bees, has there been a stage of thinking  
25 relative impact, types of measurements available, when we've got  
26 diminishing returns from better measurement, that kind of thing?



1                   **DR. THOMAS STEEGER:** I think what we're trying  
2 to depict here is the generalized process that the agency -- it  
3 plans on or proposes to use for honey bees and to the extent  
4 that honey bees serve as surrogates for non-Apis bees as well.  
5 And this process has to rely, at least initially, as with all  
6 taxa, a screening level that is relatively generic and looks at  
7 very broad categories of effects to see where resources really  
8 need to be expended to focus in on where risk, at this very  
9 broad level, cannot be dismissed.

10                   So when you talk about the nuances about  
11 colony level effects --

12                   **DR. NINA FEFFERMAN:** Forgive me. I don't  
13 actually mean nuances of particular effects; I mean actually  
14 going more abstract rather than more specific.

15                   There seems to me to be layers of very  
16 specific and then we measure this type of pesticide application  
17 and how much gets into food supply. You know, that all seems to  
18 be to me to be a very specific measurement as opposed to much  
19 broader things like, well, what are the toxicological effects at  
20 this layer, as opposed to we measure this flow path or something  
21 like that.

22                   Has that kind of assessment of a more abstract  
23 flow process for risk been accomplished already?

24                   **DR. THOMAS STEEGER:** I think -- and correct me  
25 if I'm wrong -- you're getting at this issue that the agency  
26 routinely wrestles with and it's this concept of a adverse



1 outcome pathway and how do you transition from one level of  
2 biological organization to the next.

3 I think that we're looking, as this process  
4 evolves, to help develop a more standardized approach for making  
5 those linkages. That's one of the difficulties that we have in  
6 the use of many of the sublethal endpoints that have been  
7 identified in some of the bee studies that are reported in the  
8 open literature. That's a challenge.

9 I'll say that we don't have all the answers at  
10 this point. But for the frank effects that we traditionally use  
11 of acute lethality and survival and reproduction. Those are the  
12 affects that right now are what the assessment endpoints are  
13 based on and the measurement endpoints that we plan to quantify  
14 as we develop this process further.

15 But developing those linkages that would  
16 better enable us to move from one level of biological  
17 organization to the next, you're absolutely right. That's what  
18 the ultimate endgame is. Have they been developed yet? No.  
19 But the science is still evolving.

20 **DR. NINA FEFERMAN:** Okay. Great. Thanks.  
21 So you're thinking of that as something that comes out of this  
22 level as opposed to something that is done before we get down to  
23 the drill down level of these discussions?

24 **DR. THOMAS STEEGER:** I think that the process  
25 that we're using to refine risk assessment where we identify  
26 chemicals that there appears to be risk that can't be



1 discountable at the screening level, that we move to higher tier  
2 testing at the whole colony level to try to detect whether under  
3 actual use conditions is the colony going to be able to survive,  
4 either in the short-term or in the long-term?

5 Having very bright lines at the full colony  
6 level is difficult to establish at this point because these  
7 nuances that I mentioned earlier, we don't really have a clear  
8 understanding of how much of an impact can a colony sustain or a  
9 population of bees can sustain, over long periods of time.

10 I think that that is something that still has  
11 to evolve, but our screening level process is geared towards  
12 trying to do that type of refinement, but at this point, it  
13 would be a qualitative assessment to determine is the colony  
14 going to make it or not.

15 **DR. NINA FEFFERMAN:** Thank you. I think your  
16 comments directly address my question. So I guess my take home  
17 from this might be that on some level, we might consider, as  
18 we're talking, that some of this might be putting the cart  
19 before the horse, where as if, when we get to that qualitative  
20 level, we discover that some of these aspects are not having any  
21 impact than the careful ability to measure the local impact, may  
22 not help us build up to that level. So that's great. Thank  
23 you. I just want to get that on the record. Thanks.

24 **DR. THOMAS STEEEGER:** I think you put that  
25 very well.

26 **DR. DANIEL SCHLENK:** Great. Thanks a lot



1 for getting that clarified. Okay. Our next speaker is  
2 Christina Wendel. Oh, sorry. You have a second one? Oh,  
3 sorry. Keith Sappington, please.

4 **MR. KEITH SAPPINGTON:** Thank you. In my  
5 second presentation, I will describe two risk assessment  
6 decision frameworks that we've proposed to use as guide for  
7 assessing pesticide risk to honey bees. Here, I will only  
8 summarize the proposed decision frameworks and their components  
9 and detail specific to each of these components will be  
10 presented in subsequent presentations.

11 In describing the proposed decision  
12 frameworks, I'll first begin by summarizing some of their key  
13 attributes which shape their development. I will then describe  
14 the proposed decision frameworks for pesticides that are applied  
15 via foliar sprays and then for pesticides applied via soil and  
16 seed treatment applications.

17 Lastly, I will conclude with a comparison of  
18 our proposed decision-making process with other decision  
19 frameworks that we've identified in the literature. Before  
20 describing the frameworks themselves, I think it's important to  
21 discuss some of the attributes or factors that shape their  
22 development and, as a consequence, their overall scope.

23 First of all, the agencies represented here  
24 are responsible for assessing ecological risk of over 1,000  
25 active ingredients for conventional pesticides. As a result,  
26 one attribute that shaped the proposed risk assessment process



1 for bees was the need to efficiently screen out pesticides that  
2 posed little or no potential risk concern to bees from those  
3 that have a potential risk concern.

4 To accomplish this, we are proposing a tiered  
5 process that begins with, at first, in Tier I, with a simple  
6 screening level assessment. And by design, this Tier I process  
7 is both conservative and quantitative in nature. And by that, I  
8 mean that it relies on high-end estimates of exposure and  
9 results in a derivation of a numeric risk quotient.

10 The proposed Tier I assessment considers  
11 effects measured at the individual level of biological  
12 organization, under controlled laboratory conditions. Tiers II  
13 and III involve increasing information requirements that are  
14 typically satisfied by studies conducted in the field. And as a  
15 result, Tiers II and III involve a greater environmental realism  
16 in pesticide exposure compared to Tier I.

17 In some cases, pesticide exposure information  
18 generated in Tiers II and III may be used to refine exposure  
19 estimated in Tier I. Importantly, however, in Tiers II and III,  
20 effects are evaluated at the colony level.

21 Our proposed frameworks are also shaped by the  
22 need to rely largely on toxicity studies which have established  
23 regulatory guidelines; however, we and others have noted  
24 significant gaps in the existing battery of toxicity test  
25 guidelines for bees. Therefore, these frameworks include  
26 placeholders for several additional studies for which test



1 methods have not been fully vetted as regulatory guidelines, but  
2 protocols are available.

3 The proposed decision frameworks also focus on  
4 major exposure pathways of concern to honey bees and are also  
5 shaped by the need to have sufficient information available from  
6 which a quantified pesticide exposure from these pathways.

7 For example, some exposure pathways, such as  
8 ingestion of contaminated surface water are considered less  
9 important routes of exposure for honey bees, relative to direct  
10 contact and the consumption of pollen and nectar.

11 Other pathways, such as consumption of plant  
12 guttation fluid may contain significant concentrations, but the  
13 degree to which honey bees rely on this source of water and  
14 nutrients has proven difficult to quantify.

15 And again, later this morning, Ms. Christina  
16 Wendel will provide further evaluation of the selection of  
17 exposure pathways considered in this risk assessment process.  
18 The proposed decision frameworks also differ depending on the  
19 type of pesticide application which is consistent with the  
20 conceptual models that I presented previously.

21 And lastly, decisions on moving from one tier  
22 to the next, which incorporate both risk assessment and risk  
23 management considerations are based on multiple lines of  
24 evidence and considerations of uncertainty.

25 In addition to these attributes, I also wish  
26 to make several points regarding the potential application of



1 these frameworks and future pesticide risk assessments involving  
2 honey bees. First, I'll be describing them as one moves from  
3 the top in Tier I, down to the bottom in Tiers III.

4 As we envision this top to bottom application  
5 of the frameworks, may be more typical of their use and new  
6 pesticide risk assessment where the information base is just  
7 being developed.

8 However, for existing pesticides, relevant  
9 information may be available at multiple tiers at the onset of a  
10 risk assessment, and in this case, the expectation would be that  
11 the decision-making process would reflect all available  
12 information available at the time of the assessment and may  
13 involve starting at various tiers simultaneously in the  
14 assessment.

15 The second bullet emphasizes that the decision  
16 frameworks are intended to reflect an iterative process, whereby  
17 information gained at higher tiers can and should be used to  
18 refine information and assessments conducted at the lower tiers  
19 and vice-versa.

20 Third, these decision frameworks are intended  
21 to be used as a guide to risk assessors and not as a rigid and  
22 inflexible recipe for decision-making.

23 And then finally, while the frameworks reflect  
24 current state of the sciences as we understand it, the  
25 expectation is that as information and test guidelines are  
26 developed in the future, for example, for non-Apis bees,



1 appropriate modifications will be made to the frameworks.

2 So I will first begin by stepping through the  
3 proposed decision framework for assessing risk for honey bees to  
4 foliar spray application of pesticides, and this would include  
5 systemics as well as non-systemics.

6 The first step in this process is to collect  
7 all relevant information regarding the nature of its pesticide  
8 and its use pattern, and this is a routine step in the problem  
9 formulation phase that includes gathering information on the  
10 pesticide's chemical, physical and toxicological properties, as  
11 well as detailed information on its environmental fate.  
12 Information on the timing and location and type and  
13 attractiveness of crops to which pesticides are being applied is  
14 also evaluated.

15 Shown here, the Tier I begins with assembling  
16 this information and asking the question whether or not exposure  
17 of adults or brood to the pesticide is likely to be a potential  
18 concern.

19 For outdoor spray applications, the answer  
20 would likely be yes, but for some other applications such as  
21 indoor or greenhouse uses, the answer would likely be no.

22 In cases where pesticide exposure is not  
23 considered a potential concern at this stage, a presumption of  
24 minimal risk would be made. If exposure is considered to be a  
25 concern, then three major exposure routes would be evaluated in  
26 the Tier I part of the assessment. The first of these includes



1 direct contact of adult foraging worker bees to the pesticide  
2 which intercept the spray droplets during flight.

3 We have proposed a process to derive  
4 conservative or high-end estimated environmental concentration,  
5 abbreviated EEC, to the bees via contact exposure, as depicted  
6 in Box 3(a) of this diagram.

7 The derivation of this EEC and other EECs used  
8 in Tier I will be discussed later by Ms. Kris Garber. As shown  
9 in Box 4(a), an acute risk quotient, or RQ, is then determined  
10 by the ratio of the contact EEC to the acute contact LD50.

11 The second exposure route that would be  
12 evaluated is oral exposure of adult foraging bees. And again,  
13 here a conservative estimate of exposure via consumption and  
14 nectar is calculated as shown in Box 3(b) and an acute oral RQ  
15 is determined as a ratio of this oral EEC to the acute oral LD50  
16 for adult honey bees. And that's shown in Box 4(b).

17 In addition, we propose that a measure of  
18 chronic risk to adult foragers be determined in Tier I by  
19 dividing the same EEC by the no-observed adverse effect  
20 concentration, or NOAEC, from a chronic toxicity study on adult  
21 bees. As noted with the asterisks, a regulatory guideline is  
22 not yet available for this study, although various protocols  
23 have been described in the literature.

24 In the third exposure pathway proposed for  
25 evaluation in Tier I is oral exposure to developing honey bee  
26 brood. An oral EEC for larvae would be determined, as shown in



1 Box 3(c) and the acute and chronic oral risk quotient values  
2 would be calculated using acute oral LD50 and a chronic oral  
3 NOAEC from the larval toxicity test.

4 We note again that the regulatory guidelines  
5 have yet to be developed or finalized for measuring larval  
6 toxicity in the laboratory, and these would need to go through  
7 the formal guideline development process.

8 Once these RQ values are determined, they will  
9 be compared to the proposed level of concern, abbreviated LOC.  
10 The proposed LOC for evaluating acute risk is 0.4, and that  
11 proposed for evaluating chronic risk is 1.0. The technical  
12 basis of these LOC values will be described later by Dr.  
13 Steeger.

14 As the LOC comparisons are used to inform the  
15 decision to proceed with additional refinement or move on to  
16 Tier II; however, not explicitly depicted in Box 5, is that this  
17 decision would not only include a comparison to the LOC values,  
18 but also other lines of evidence, including available ecological  
19 incident reports, pesticide mode of action and uncertainty in  
20 the available information base.

21 After considering the LOC, other lines of  
22 evidence, if no significant risk is indicated then a presumption  
23 of minimal risk would be made and documented. If risk could not  
24 be ruled out at this Tier I screening phase, then other  
25 information could be considered for refining the exposure  
26 estimate, which may include residue information derived from



1 crop magnitude of residue studies which are routinely submitted  
2 to the Agency. Additional detail on this refinement step will  
3 be provided by Mr. Baris in a subsequent presentation.

4 If the applicable and refined RQ value is  
5 recalculated using this refined information and other lines of  
6 evidence are considered, the decision then becomes, again,  
7 whether to presume minimal risk or continue with the risk  
8 assessment process. As shown in Box 8, the next step involves  
9 considering the available risk mitigation options and  
10 uncertainties in the assessment and other lines of evidence as  
11 part of this decision to move on to Tier II.

12 The risk mitigation option, such as modifying  
13 the pesticide application rates, timing of application, types of  
14 crops being treated, could then be evaluated by proceeding back  
15 through the Tier I process, as indicated by this dashed black  
16 arrow on the right of the diagram.

17 If after weighing this information, including  
18 a dialogue with risk managers, information from Tier II studies  
19 is determined to be necessary, then different types of studies  
20 could then be considered.

21 One type of study may be an exposure-based  
22 field or semi-field residue study in which pesticide residues in  
23 pollen and nectar are quantified using pesticide applied to a  
24 treated crop. Information from this study could then be used to  
25 refine the estimates of pesticide exposure from pollen and  
26 nectar used earlier in Tier I.



Another type of study would be considered in Tier II as a semi-field effects-based study, such as a tunnel enclosure or high feeding study. Semi-field studies provide pesticide effects information gathered at the whole hive level. And endpoints from these studies may include, but are not limited to worker and brood mortality, brood development, foraging activity, behavioral observation, colony strength and survival. Additional information on the Tier II semi-field studies will be provided by Mr. DeCant Later this afternoon.

Once this information is gathered from the Tier II studies, other lines of evidence are considered as well as the uncertainties and potential risk to honey bees is again assessed. The outcome of this assessment may be a presumption of minimal risk, or if risk is identified, available risk mitigation options would be considered and evaluated, as indicated in Box 11.

Although not shown here, evaluation of these mitigation options might also involve preceding back to the beginning of the process. After evaluating these risk mitigation option and considering, again, all the lines of evidence and uncertainties, a decision would be made to proceed to Tier III.

And Tier III, as we've depicted it, includes full field studies that are designed to address specific uncertainties raised in the earlier tiers. An example of this might be the design and conduct of a field study specifically to



1 track overwintering success of colonies following pesticide  
2 exposure.

3 As shown in Box 13, the results of the Tier  
4 III field studies are then evaluated and available risk  
5 mitigation options are considered, and again, along with other  
6 lines of evidence and uncertainties. Risk associated with these  
7 mitigation options may be evaluated, as indicated by this black  
8 arrow, and risk is then characterized.

9 This risk characterization may lead to a  
10 presumption of minimal risk or it may lead to a presumption of  
11 risk from the Tier III studies, as well as the other earlier  
12 information. Dr. Tom Steeger will be providing additional  
13 information on this risk characterization step this afternoon.

14 The proposed decision framework for risk  
15 assessment of soil application and seed treatment application  
16 begins in a similar way that is depicted for foliar application.

17 That is, information on the pesticide and the nature of its  
18 application are evaluated and questions are asked whether or not  
19 exposure is a potential concern to adults or brood.

20 If there are little or no potential exposure  
21 of adults or brood, then a presumption of minimal risk is made.  
22 But if there is potential for exposure, then conservative  
23 estimates of oral exposure to adult worker bees and larvae are  
24 then determined, as indicated in Boxes 3(a) and 3(b) of this  
25 figure.

26 As noted in Boxes 4(a) and 4(b), Tier I



1 screening level, acute and chronic risk quotients are then  
2 calculated using these EECs and the relevant toxicity endpoints  
3 from the acute and chronic oral toxicity studies to adults and  
4 larvae are considered.

5 As noted in the conceptual model and in the  
6 White Paper for seed treatments, exposure to bees from seed coat  
7 dust has been shown to be an important route of concern and the  
8 focus of numerous ecological incident reports.

9 At this time, we're not proposing a risk  
10 assessment process for this exposure pathway, primarily because  
11 methods for predicting a priori pesticide exposure from drift of  
12 seed coat dust are not yet available for regulatory application  
13 and because the Agency is currently working to mitigate this  
14 exposure pathway based on modifications of seed treatment  
15 methods, seed planting equipment and associated practices.

16 In addition, if there's a potential exposure  
17 for offsite drift of spray applications of soil, this would be  
18 addressed, as described previously, for the foliar spray  
19 applications after accounting for the fraction of pesticide that  
20 is subject to drift.

21 As shown in Box 5, the acute and chronic risk  
22 quotients are then compared against the same LOC values as  
23 described previously. These RQ values are then considered along  
24 with other lines of evidence, such as incident reports, to  
25 determine whether a presumption of minimal risk can be made or  
26 if additional refinements are considered necessary in the Tier I



1 exposure estimate.

2 The remainder of the proposed decision  
3 framework is the same as I described for foliar sprays.  
4 However, the design of tests in Tiers II and III would reflect  
5 the specific pesticide application methods being used. And the  
6 EECs calculated in Tier I would also differ in their method.

7 Lastly, we thought it would be useful to place  
8 the proposed decision framework I just presented in the context  
9 of two recently published risk assessment schemes for bees.

10 The first is presented in a scientific opinion  
11 document developed by the European Food Safety Authority, or  
12 EFSA, earlier this year. And the second is a decision scheme  
13 that was published by the Society of Environmental Toxicology  
14 and Chemistry, SETAC, in 2011, and based on an international  
15 Pellston-style Workshop. These efforts were very useful to us  
16 as we developed the White Paper.

17 And as a result, we note a number of  
18 similarities among the three assessment schemes. First of all,  
19 as indicated in the first bullet, they all involve a tiered risk  
20 assessment process that begins with assessing effects at the  
21 individual level, based mostly on laboratory studies. This is  
22 then followed by effects which are assessed at the colony level  
23 at higher tiers.

24 As indicated in the second bullet, they all  
25 contain risk assessment schemes that are tailored to different  
26 application methods; for example, sprays, soil and seed



1 treatment.

2 Third, exposure to pesticides from contact  
3 routes as well as oral exposure is prominent in all three  
4 schemes, although the EFSA scheme does include a conservative  
5 estimate of dust exposure from seed treatments as a fraction of  
6 the overall calculated application rate. It's not clear at this  
7 time which methods would then be used to refine this screening  
8 level estimate.

9 Fourth, all schemes recognize the need to  
10 assess both acute and chronic toxicity early in the risk  
11 assessment process. I wish to point out some of the differences  
12 that we've noted as well. First, one example is that the  
13 triggers used to move from Tier I to higher tiers differ in  
14 their form.

15 The process proposed in the White Paper bases  
16 the Tier I trigger value on the ratio of the estimated dose to  
17 the corresponding toxicity value. The approach recommended in  
18 the EFSA Scientific Opinion document, bases its trigger on the  
19 ratio of the application rate in grams per hectare to the  
20 toxicity value.

21 As application rate is obviously related to  
22 pesticide doses, the two approaches are similar in concept, but  
23 they differ in the details. Another difference is that non-Apis  
24 bees are explicitly included in the risk assessment schemes  
25 recommended by the SETAC Pellston Workshop and EFSA. However,  
26 these schemes acknowledge that additional test guideline



1 development is needed before they can be fully implemented for  
2 non-Apis bees.

3 Lastly, the proposed approach in the White  
4 Paper recommends characterizing risk by considering the overall  
5 weight of evidence at higher tiers, which would include results  
6 from semi-field and field studies, but also would include other  
7 lines of evidence provided by suitable studies from the  
8 literature and incident reports. It appears to us that the risk  
9 conclusions at higher tiers from these other schemes are largely  
10 limited to the results from the applicable semi-field and field  
11 studies.

12 I want to thank you very much for your  
13 attention. At this time, myself or the team would be pleased to  
14 address your questions on the overall risk assessment process,  
15 noting that detailed questions related to components of this  
16 process are probably best reserved for the specific presentation  
17 this afternoon. Thank you.

18 **DR. DANIEL SCHLENK:** Any questions? Dr.  
19 Berenbaum.

20 **DR. MAY BERENBAUM:** This may be best reserved  
21 for the afternoon, but just as a point of clarification, the  
22 acute and chronic toxicity testing at Tier I is described in  
23 discrete life stages. Is there any consideration of a way to  
24 incorporate life cycle testing rather than just individual  
25 discreet stage testing, sort of risk, kind of concept?

26 **MR. KEITH SAPPINGTON:** In Tier I, we've only



1 described, as you say, the larval and the adult stages. The  
2 larval toxicity test, as I understand, that we are evaluating is  
3 one that could continue out through emergence, but at this time,  
4 it's quite challenging to get acceptable responses in the  
5 controls from this.

6 But I would believe with additional  
7 refinement, we would be able to perhaps capture that part of the  
8 development cycle. For capturing the entire life cycle, I  
9 believe those are reserved now in Tier II, where we would be  
10 looking at the hive level.

11 **DR. DANIEL SCHLENK:** Dr. Potter.

12 **DR. THOMAS POTTER:** Yes. Just a quick  
13 question about the LOC values that you proposed. Would those be  
14 bright line values in a sense that the value is fixed as .4 and  
15 you have a chemical that is say, .41? You know, what's the  
16 process there?

17 **DR. THOMAS SEEGER:** When a compound or an RQ  
18 value exceeds an LOC, that information is relayed back to the  
19 risk manager. The decision to transition to higher tier levels  
20 or the degree to which the LOC is exceeded, that's a risk  
21 management decision.

22 So would you consider it a bright line? It is  
23 a bright line in the sense that the LOC is fixed for screening  
24 level assessment. Whether it triggers the need to do higher  
25 tier assessments is a risk management decision.

26 **DR. DANIEL SCHLENK:** Dr. Bradbury, you want



1 to add a little bit more to that?

2 **DR. STEVEN BRADBURY:** That LOC isn't provided  
3 in isolation from other information. So your .41 versus .39,  
4 what we want to know is what do we know about the mode of  
5 action; what does the dose response curve look like?

6 All that information that Keith's been  
7 describing in the conceptual model would all come into play in  
8 terms of making a decision about what the uncertainties are and  
9 what would be the magnitude and the nature of the uncertainties  
10 around that threshold.

11 But Dr. Steeger is right in that that LOC  
12 value is a value we use to make sure we pause, let's think,  
13 let's talk, let's look at all the information we got and make  
14 some decisions about what the next most logical step would be.

15 **DR. DANIEL SCHLENK:** Okay. Dr. McManaman.

16  
17 **DR. JAMES MCMANAMAN:** Is it the Agency's view  
18 that the LD50 is the most conservative method estimate of  
19 toxicity for larvae or have you considered other possibilities?

20 **DR. THOMAS STEEGER:** The LD50 and the LC50 are  
21 the traditional measures that the Agency relies on for screening  
22 level because it's considered the most precise estimate from the  
23 acute toxicity studies where the confidence limits around that  
24 estimate are at their narrowest point.

25 **DR. JAMES MCMANAMAN:** But it may be the most  
26 reliable, but you view it as the most conservative estimate,



1 especially for this first tier of screening?

2 **DR. THOMAS STEEGER:** Whether it's the most  
3 conservative value, I mean, certainly, there are more  
4 conservative values that could be selected, but again, the  
5 precision that's surrounds those, the confidence limits that  
6 would be surrounding those estimates would be highly variable.  
7 At this point, that's the traditional measure that we, as a  
8 regulatory authority, as well as other regulatory authorities  
9 have used as a conservative measure of acute toxicity.

10 **UNIDENTIFIED SPEAKER:** I wondered the same  
11 thing, actually.

12 **DR. DANIEL SCHLENK:** Excuse me. Somebody  
13 on the telephone, can you please mute your line again, please?  
14 Thank you.

15 **MR. KEITH SAPPINGTON:** I'd like to follow-up  
16 on that question. Again, I agree that the endpoint selected  
17 from the acute studies, such as the LC50 and the LD50 is the  
18 most precise estimate, but we then apply this level of concern  
19 to that. Dr. Steeger will be presenting the basis for that, so  
20 I don't want to get into that in significant detail. Part of  
21 the basis is the extrapolation down to a lower effect level, in  
22 this case, a ten percent effect level.

23 **DR. DANIEL SCHLENK:** Dr. Bradbury. I saw  
24 your hand up. Did you want to --

25 **DR. STEVEN BRADBURY:** Keith beat me to the  
26 punch. The concept is a dose response relationship factors into



1 that LOC. So it's a different way to get at the concept, but I  
2 think in the talks this afternoon it'll come out in more detail.

3 **DR. DANIEL SCHLENK:** Dr. McManaman.

4 **DR. JAMES MCMANAMAN:** I understand the  
5 concepts here, but whereas with adult bees I can see that it  
6 might be the appropriate measurement, but I'm concerned that for  
7 larvae, it may not be the appropriate measurements. So that was  
8 the basis of my question.

9 **DR. DANIEL SCHLENK:** Okay. Dr. Pettis.

10 **DR. JEFF PETTIS:** Just a point of  
11 clarification about, currently the White Paper is considered  
12 honey bees as a surrogate for all pollinators. And there's  
13 little consideration of the other life history for solitary  
14 bees; is that correct?

15 **DR. THOMAS STEEGER:** You're correct that  
16 the proposed framework is using the honey bee as the focus and  
17 we are asking the SAP to comment on the extent to which it can  
18 be applied to non-Apis bees as well. But those measures of  
19 effect are based on individual bees which would presumably  
20 relate to solitary native bees.

21 And at higher tier assessments, you're looking  
22 at colony level effects that would presumably speak to potential  
23 effects to social and non-Apis bees and as will be pointed out,  
24 and was already pointed out in preliminary presentations, but  
25 will be further discussed by Kris Garber, the measures of  
26 exposure are through diet and through contact which would



1 presumably be relevant to ground nesting bees, solitary bees or  
2 non-Apis bees, just as it is relevant to the honey bee.

3 **DR. DANIEL SCHLENK:** That was Dr. Steeger.

4 Any other questions about the framework?

5 (No response.)

6 Okay. So now I think we're ready for Ms.  
7 Wendel, is that right?

8 **MS. CHRISTINA WENDEL:** Hello. My name is  
9 Christina Wendel and I'm a biologist in the Environmental Fate  
10 and Effects Division of the Office of Pesticide Programs. And  
11 thank you for the opportunity to address the panel.

12 This morning I will be talking about the  
13 exposure of honey bees to pesticides, the exposure routes that  
14 were considered and their overall relative importance in terms  
15 of major versus minor routes of exposure.

16 Discussion of the potential exposure pathways  
17 is presented in the White Paper as part of the conceptual model  
18 in Section II, the Tier I exposure assessment in Section III,  
19 and the drinking water consumption rate analysis is in Appendix  
20 Number 2. This presentation relates to Charge Question Number  
21 7.

22 I will be presenting the exposure routes  
23 identified and considered in the conceptual model as presented  
24 and described earlier by Mr. Sappington. I will also briefly  
25 discuss and describe the rationale used in determining the  
26 exposure routes identified for use in the Tier I exposure



1 assessment, as well as those routes that were not included.

2 Ms. Garber will follow this presentation with  
3 a more in-depth analysis of our proposed Tier I exposure  
4 assessment and the primary routes of exposure that were chosen  
5 and quantified.

6 For the Tier I exposure assessments, three  
7 types of exposure routes were considered. These are contact,  
8 oral, and inhalation exposure. As identified previously,  
9 exposure routes differ with the type of application, which may  
10 include foliar sprays, soil applications, and seed treatments.

11 In addition, exposure routes also differ based  
12 on the type of bee. Exposures differ for larval and adult bees  
13 and for the castes of the bee, which include workers, drones,  
14 and queen bees.

15 The next few slides provide visual  
16 representations of exposure routes for bees based on both the  
17 application methodology and the type of bee. Again, this slide  
18 provides a visual representation of a honey bee and depicts the  
19 potential exposure routes for forager bees for foliar spray  
20 applications.

21 For foliar spray applications, adult forager  
22 worker bees have the greatest potential for exposure as they  
23 leave the hive. Forager bees have direct contact with  
24 foliarly-applied pesticides via direct spray and contact with  
25 pesticide residues on foliage and soil.

26 Forager bees also are potentially exposed to



1 pesticide residues via the collection of pollen, nectar,  
2 drinking water and plant resins or propolis that may be brought  
3 back to the hive. Another route of exposure for forager bees  
4 includes oral exposure through the diet from the consumption of  
5 contaminated pollen, nectar and/or drinking water.

6 It is important to note that it was assumed in  
7 the Tier I exposure method that the major route of oral exposure  
8 is through the actual consumption of food. However, there could  
9 potentially be a dose of exposure for the forager bee from the  
10 carrying transport of pollen, nectar, and/or drinking water back  
11 to the hive. But we expect that the dose obtained from the  
12 consumption of food would be greater and productive of any dose  
13 obtained during the transport of pollen, nectar, and/or drinking  
14 water to the hive.

15 Therefore, the oral exposure obtained from  
16 diet and not during the collecting phase was considered to be  
17 the oral route of exposure. During flight, forager bees may  
18 also be exposed via inhalation of spray droplets and/or the  
19 gassiest phase of volatilized chemicals.

20 This slide provides a visual representation of  
21 the honey bee and depicts the potential exposure routes for hive  
22 bees for foliar spray applications. Adult bees living within  
23 the hive include workers, for example, nurse bees, as well as  
24 drones and queen bees.

25 The adult hive bees are exposed primarily  
26 through the oral route of exposure from the consumption and



1 processing of pollen and nectar into bee bread, honey, brood  
2 food and/or royal jelly, or the consumption of drinking water.

3 There is no longer contact exposure to direct  
4 spray; however, contact exposure can occur within the hive  
5 through the transfer of pollen and/or nectar from forager bees  
6 to the hive worker bees, as well as direct contact with comb wax  
7 and propolis.

8 Queen bees can also be exposed to direct spray  
9 and inhalation spray of spray droplets during mating and  
10 orientation flights; however, the dominate exposure route for  
11 queen bees is based on the exposure route as she consumes royal  
12 jelly throughout her life in the hive.

13 For larval bees, items must be brought back to  
14 the hive and processed prior to larval exposure. Larval bees  
15 are exposed to the ingestion of the processed pollen and nectar,  
16 as royal jelly and/or brood food. As is the case for all adult  
17 and larval bees within the hive, inhalation of the gassiest  
18 phase of a volatilized chemical is also a potential exposure  
19 route.

20 This slide also provides a visual  
21 representation of the honey bee and depicts the potential expose  
22 routes for forager and hive bees for soil and seed treatment  
23 applications. For soil and seed treatment applications, there  
24 is no longer contact exposure to direct spray. Instead, for  
25 seed treatment applications, there is potential contact exposure  
26 resulting from seed coat dust abrasion.



1 In addition, for both adult and larval bees,  
2 oral exposure through the diet remains a major route of exposure  
3 for soil and seed treatment applications. For systemic  
4 compounds, translocation and route uptake occurs, resulting in a  
5 transfer of a chemical into the growing plant tissues and  
6 exudates, including pollen, nectar, as well as drinking water.

7 Although the magnitude of exposure may be  
8 different between forager bees and hive bees, there is the  
9 potential for contaminants to be brought back to the hive and  
10 transferred to the bees living within the hive, based on  
11 grooming, the transfer of the collected items to the hive bees,  
12 the processing of pollen and/or nectar into bee bread, honey,  
13 brood food and/or royal jelly, the ingestion of the processed  
14 pollen and nectar products, as well as direct contact with comb  
15 wax and/or propolis.

16 Again, for both adult and larval bees,  
17 inhalation is a potential exposure route in the hive from  
18 exposure to the gassiest phase of a volatilized chemical.

19 During the Tier I exposure assessment, the  
20 exposure routes presented within the conceptual models for all  
21 application methodologies and chemical classes, including  
22 systemic and non-systemic compounds were examined and  
23 considered.

24 Based on our review of the available  
25 information, the major routes of exposure of bees to pesticides  
26 are contact from direct spray for foliar spray application and



1 oral exposure from the ingestion of the contaminated pollen and  
2 nectar for all pesticide application methods.

3 The primary routes of exposure are shown here  
4 in green. These two routes of exposure are quantified in the  
5 Tier I exposure assessment by generating estimated environmental  
6 exposure concentrations, EECs. The process used to validate the  
7 Tier I exposure method will be discussed in the next  
8 presentation.

9 For contact exposure, direct spray is the most  
10 conservative exposure route for forager bees and covers the  
11 other potential contact exposure routes shown here in blue,  
12 including contact with contaminated foliage, soil, comb wax,  
13 propolis and/or pollen.

14 For oral exposure, the ingestion of  
15 contaminated pollen and/or nectar are assumed to be protective  
16 of other food consumed by bees, shown here in blue, and include  
17 honey, bee-bread, royal jelly, and brood food.

18 Contact exposure via dust, oral exposure  
19 resulting from consumption of contaminated drinking water and  
20 inhalation exposure from spray droplets and/or the gassiest  
21 phase of a volatilized chemical, shown here in red, were  
22 considered and evaluated separately and will be discussed in the  
23 next several slides.

24 Contact exposure from abraded dust as a result  
25 of seed treatment application is a potential major route of  
26 exposure to bees. Numerous incidents related to abraded dust



1 from seed treatment applications have been reported in North  
2 America. As a result, the agencies are actively working with  
3 registrants, growers, applicators and seed equipment  
4 manufacturers to minimize this exposure pathway. Different  
5 technologies, stewardship programs and a variety of different  
6 mitigation measures, including label language, have been  
7 developed and research in these areas continues.

8 Mitigation measures have been identified,  
9 including BMPs, new seed coat technologies, and alternative low  
10 dust lubricating agents to replace high dust producing  
11 lubricants like talc and graphite. In addition, best management  
12 practices are being considered and developed. A review of these  
13 processes will continue to occur as these technologies and  
14 practices advance over time.

15 Although dust is considered to be a major  
16 exposure route, efforts are actively being made to minimize this  
17 route of exposure to bees. As a result, exposure to dust was  
18 not included in the Tier I screen for all pesticides. This  
19 issue is associated only with seed treatment applications and as  
20 a result, will be dealt with on a case-by-case basis for  
21 relevant chemical and application types.

22 Water is collected for two primary reasons.  
23 First, to regulate the temperature within the hive through  
24 evaporative cooling to maintain conditions for healthy brood,  
25 and second, to use in preparing larval brood food by nurse bees.  
26



1 Water is not stored within the hive; rather,  
2 it must be collected by foraging bees when needed by the colony.

3 To reiterate, it is assumed in the Tier I Exposure Method that  
4 the major route of oral exposure is through the actual  
5 consumption of food, recognizing that there could potentially be  
6 a dose of exposure for the forager bee from the carrying and  
7 transport of the pollen nectar and/or drinking water back to the  
8 hive. But again, we expect that the dose obtained through the  
9 consumption of food would be greater and protective of any dose  
10 obtained through the transport of these materials.

11 EPA completed an analysis to estimate  
12 potential exposure doses bees could receive from consuming  
13 contaminated drinking water. This analysis is presented in  
14 detail in Appendix 2 of the White Paper. Based on estimated  
15 pesticide concentrations in various sources of water potentially  
16 consumed by bees and water flux rate from the brown paper wasp,  
17 the results of this analysis indicate that if bees consume the  
18 majority of their water from ponds, including water adjacent to  
19 treated fields or puddles of water located on treated fields,  
20 the exposure is relative to dietary and direct spray are  
21 discountable.

22 However, if bees drink a substantial amount of  
23 water from guttation fluid found in crops, or dew, the  
24 condensation on the plant, with exposure from dew occurring on  
25 crops foliarly sprayed with pesticides, conservative exposures  
26 may be similar to or even exceed pesticide exposures through the



1 diet or direct spray. Given the potential concern associated  
2 with exposure through drinking dew and guttation fluid, these  
3 exposure pathways were investigated further.

4 Based on the results of EPA's analysis  
5 presented in Appendix 2 of the White Paper, pesticide exposures  
6 through dew and guttation fluid are not expected to be as  
7 significant when compared to the diet and direct spray because  
8 of two primary reasons. First, the relative importance of dew  
9 and guttation fluid as a source of drinking water to bees is  
10 unknown. In addition, dew and guttation fluid are expected to  
11 be present only during a portion of the day, typically in the  
12 morning, with a very conservative maximum of six hours overlap  
13 in foraging times of bees and the presence of dew and/or  
14 guttation fluid, preventing bees from drinking a substantial  
15 amount of water from these sources.

16 Secondly, for many worker bees, pesticide  
17 doses, through the consumption of contaminated dew and/or  
18 guttation fluid, may be much smaller due to lower or  
19 non-existent drinking water consumption rates when compared to  
20 the percentage of water consumed from food.

21 As shown in this slide, the estimated food  
22 consumption rates and resulting percentage of water consumed as  
23 part of food, including honey or nectar for adult worker and  
24 forager bees, ranges from 7 percent to greater than 100 percent  
25 of their daily water requirement. And again, these calculations  
26 are presented in Appendix 2 of the White Paper.



1                   Therefore, for many bees, pesticide exposure,  
2 through the consumption of either dew and/or guttation fluid is  
3 expected to be minimal because of higher amounts of water are  
4 likely consumed through food rather than from drinking water.  
5 Therefore, based on this analysis, pesticide exposures, through  
6 drinking water, was not included in the proposed Tier I exposure  
7 method for bees.

8                   Inhalation is a relevant exposure pathway for  
9 only highly volatile chemicals, including fumigants applied via  
10 foliar or soil application, where hives are in close proximity  
11 to the treated site. It is not a relevant exposure route for  
12 seed treatment applications.

13                   Based on a preliminary analysis conducted  
14 after the White Paper was developed, it was determined that  
15 exposure to bees via the inhalation route is substantially lower  
16 than the dietary exposure routes. Therefore, the inhalation of  
17 exposure pathway was not included as an exposure route in the  
18 proposed Tier I Exposure Method for bees.

19                   In summary, the primary routes of exposure  
20 that are quantified in the proposed Tier I method include  
21 contact resulting from direct spray and oral exposure from the  
22 dietary consumption of pollen and nectar. Although there are  
23 other potential exposure routes, these were evaluated and found  
24 to be minor in comparison to the direct contact and oral routes.

25  
26                   Therefore, these other routes of exposure were



1 not included within the proposed Tier I Exposure Assessment. We  
2 are seeking the input from the SAP on the proposed contact and  
3 oral exposure routes included in the Tier I Exposure Method, as  
4 well as those exposure routes that were not included in the Tier  
5 I Exposure Method for Bees. Thank you for time. The team and I  
6 can address any questions you may have.

7 **DR. DANIEL SCHLENK:** Thank you. Any  
8 questions? Dr. James.

9 **DR. ROSALIND JAMES:** Thank you. I thought the  
10 analysis you did was very good, but I have one question. You  
11 don't seem to consider Layer I, discussing about doing the  
12 bioassays for Tier I test, combining the oral and dermal  
13 together, whereas most bees are going to get both an oral  
14 exposure and the contact exposure. What's the reason for that?  
15 Or did I misunderstand that?

16 **MS. KRISTINA GARBER:** You didn't misunderstand  
17 that. We're in the proposed process as it's depicted in the  
18 White Paper. We would just treat those two routes separately.  
19 Potentially, we could add risk quotients.

20 So one approach could be to add risk quotients  
21 of the two pathways and then compare them to an LOC. But we are  
22 in the proposed process of treating them separately and the test  
23 would be conducted separately as well.

24 **DR. THOMAS SEEGER:** I would like to add to  
25 Chris's comment. The in vitro assay that's been proposed for  
26 larval toxicity testing, those larvae are being exposed sort of



1 as a combination of both oral and contact exposure. So in a way  
2 that concern is accounted for by that assay.

3 **DR. DANIEL SCHLENK:** Okay. Dr. Tarpy.

4 **DR. DAVID TARPY:** You had two main routes of  
5 water that bees would forage on, one is ponds or kind of runoff,  
6 and the other would be dew. Where would things like irrigation  
7 systems that are often used and bees use as an important source  
8 of water that are also conduits for pesticide treatment, where  
9 would that fall among those two routes of exposure?

10 **MS. KRISTINA GARBER:** I think we weren't  
11 actually accounting for the irrigation or even direct  
12 applications to irrigation canals. That really doesn't fit into  
13 the concept of the pond. If it's a direct application or a  
14 chemigation application, perhaps, if that's what -- okay.  
15 You're nodding "yes."

16 I think that, you know, one way to deal with  
17 that could potentially be to take the labeled application rate  
18 as it appears, you know, for the chemigation application and use  
19 the drinking water ingestion rates that we've got in the paper  
20 and see if the assumptions that we've got presented here still  
21 hold. And if so, we could proceed as usual. But this kind of  
22 speaks to what Keith was talking about earlier about how our  
23 conceptual diagrams that we're presenting are kind of based on a  
24 suite of application methods that are pretty typical of  
25 agriculture.

26 In this case, I mean, chemigation is a typical



1 application as well. We could modify that conceptual model to  
2 account for this other scenario.

3 **DR. DANIEL SCHLENK:** Dr. Pistorius.

4 **DR. JENS PISTORIUS:** I have a question. You  
5 said the effective quantitative screening tool is not currently  
6 available for dust. If such methods are available, could you  
7 imagine incorporating those in the risk assessment scheme? I  
8 hope this is appropriate to ask, how long do you think it would  
9 take?

10 The second is it is a technical issue also.  
11 So it is a question of risk assessment and risk management. If  
12 such risk management measures would be possible, do you think it  
13 is appropriate to take it out from the risk assessment  
14 completely?

15 **DR. THOMAS STEEGER:** The issue of dust,  
16 abraded dust from treated seed coat, as we understand it,  
17 requires a number of factors. And the difficulty in quantifying  
18 the level of dust that could be generated that would allow us to  
19 predict it on a regular basis, we don't know if at this time  
20 such a method of prediction is possible.

21 If one were developed, then yes, I think we  
22 would consider it, but at this point we're hoping that through  
23 best management practices, the likelihood of abraded dust can be  
24 reduced such that it won't represent major route of exposure.  
25 But clearly there are incidents that suggest -- as was evidenced  
26 in Germany in 2008 -- that abraded dust can serve as a source of



1 exposure, given an alignment of factors that influenced the  
2 generation and movement of those dust from treated seed.

3 The issue of management, as Mr. Sappington  
4 portrayed throughout the movement of the framework, risk  
5 assessment or risk management options are considered throughout  
6 the process. And if we can discount potential exposure through  
7 mitigation, that would be factored into the assessment.

8 That's why the importance of maintaining close  
9 communication with the risk manager from the very start of the  
10 assessment process would enable us to better account for  
11 mitigation options that might be considered.

12 **DR. DANIEL SCHLENK:** Do you have a follow  
13 up with that Mr. Pistorius?

14 **MR. JENS PISTORIUS:** Just a second question.  
15 With the risk mitigation measures, this would also maybe be  
16 possible to imply, for instance, if there's a risk for close set  
17 up of hives near a field where there is guttation very likely.  
18 Would it be possible to make a link between risk assessment and  
19 risk mitigation measures there and apply with mitigation  
20 measures?

21 **DR. THOMAS STEEGER:** The regulating pesticides  
22 at a nation level to account for the position of colonies near  
23 crops that may be producing guttation fluid, we still -- and we  
24 welcome panel input on this -- we're still uncertain as to the  
25 degree that bees would be attracted to plants when guttation  
26 fluids are actually being expressed from plants.



1                   If we had a better understanding of that route  
2 of exposure, I think we would be in a better position to  
3 consider whether it should be included as a routine route of  
4 exposure and whether mitigation could be consistently applied to  
5 that as a way of resolving it.

6                   **DR. DANIEL SCHLENK:**       Dr. Potter.

7                   **DR. THOMAS POTTER:** I have two questions, if I  
8 may. The first has to do with the -- it's a question and a  
9 comment and it has to do with the drinking water exposure.

10                  In reading through Appendix 2, I was struck by  
11 what appears to be a considerable uncertainty in terms of the  
12 quantity of water that a bee may consume during the day. I  
13 looked at your assessment; perhaps that's all the information  
14 that's available. Not being a bee expert, I can't comment on  
15 that one way or the other.

16                  I will note, in that context of drinking water  
17 exposure that I looked at your calculations with regard to  
18 concentrations of pesticides in puddles. I think there was an  
19 error in units in application of the formula that you used, that  
20 you need to take a careful look at.

21                  My question connected to that is in looking at  
22 the possibility of pesticide contamination of infield puddles,  
23 did you look back at the work that was done in establishing the  
24 ECO Framework? It was about 10 years ago that ECOFRAM was on  
25 the scene.

26                  Buried in there, and apparently the



1 information is still available on the website, there is a very  
2 nice little model that was developed to actually look at the  
3 possibility of residues accumulating in puddles infield. I  
4 think that that might be something that you'd want to take a  
5 look at.

6 So my question for you is, did you look at  
7 that ECO Framework at all in the context of your puddle  
8 assessment?

9 **MS. KRISTINA GARBER:** No. We didn't look at  
10 the ECOFRAM report specifically for this method, but we can  
11 certainly do that. Thank you.

12 **DR. THOMAS POTTER:** My second question is with  
13 regard to dust exposure, did you consider any other dust in the  
14 bees' environment that may contain pesticide residues? And this  
15 could include windblown soil, dust that is generated from soil  
16 or sprays that have been applied. It could indeed include a  
17 dust application or formulation that contains the active  
18 ingredient.

19 My question for you is, what other forms of  
20 dust in the environment were looked at other than the  
21 descriptions you made about dust that may have been generated  
22 from abraded seed?

23 **DR. THOMAS STEEGER:** The dust that was  
24 considered was that that was abraded from treated seed because  
25 it would be the most concentrated form of dust where the active  
26 ingredient would be present. So the dust that's potentially



1 contributed through the movement of seeding equipment,  
2 disrupting soil or any type of tillage was not considered in our  
3 assessment. Again, the concentration, that would be considered  
4 from dust as a result of abraded seed coat would expected to be  
5 higher.

6 **DR. THOMAS POTTER:** I agree it would be  
7 higher, but there may be some exposure there. We'll get into  
8 that, I think, when we talk about some of the questions. At  
9 least from my rationale, I think it might be worth taking a look  
10 at.

11 **DR. DANIEL SCHLENK:** Any other questions?

12 (No response.)

13 Okay. According to my schedule and agenda, we  
14 have a lunch break scheduled. So let's go ahead and do that and  
15 be back at 12:45 and we'll resume at that time. Thanks.

16 (Whereupon, at 11:41 a.m., a  
17 luncheon recess was taken.)

18 \* \* \* \* \*

19 A F T E R N O O N S E S S I O N

20 (12:50 p.m.)

21 **DR. DANIEL SCHLENK:** Now that our permanent  
22 panel members are here, we can kind of go forward. Let's go  
23 ahead and get started.

24 Before we get started, I believe you have some  
25 answers to Dr. Potter's question.

26 **MS. KRISTINA GARBER:** Thank you. Dr. Potter,



1 I did a little research on my lunch break and I just wanted to  
2 get back to you on the puddle comments that you had made.

3 So first off, you had something about a unit  
4 error. And I was just hoping that you could note that in your  
5 written comments so that we can make sure we go back and take a  
6 look at that.

7 **DR. THOMAS POTTER:** I absolutely intend to  
8 comment on that. I just wanted to point that out, you know, I  
9 went back and ran the numbers and got something different. So I  
10 will certainly put it in the record.

11 **MS. KRISTINA GARBER:** Thank you. And then the  
12 other thing related to the ECOFRAM approach, it's my  
13 understanding that that's kind of based on a modification of the  
14 prism model. So we did actually consider that in the  
15 development of the model that we presented for estimating  
16 pesticide concentrations in puddles.

17 And we chose to go with the model that we did  
18 just simply because it was a more simple approach, you know,  
19 using the simple equilibrium-based approach, it indicated that  
20 concentrations in puddle weren't a problem. So we didn't see  
21 the need to move further to a more refined approach. That's  
22 basically why we chose to proceed the way that we did.

23 **DR. DANIEL SCHLENK:** Thanks. Okay. I  
24 guess you're up, Kristina. So go ahead and lead us on our next  
25 presentation, please.

26 **MS. KRISTINA GARBER:** Great. Thank you, Dr.



1 Schlenk. Thank you, members of the panel for allowing me to  
2 present on the Tier I Method for Estimating Exposure of Honey  
3 Bees to Pesticides. As a formal introduction, I'm senior  
4 biologist in the Environmental Fate and Effects Division of the  
5 Office of Pesticide Programs.

6 Now, the material that I'm going to cover  
7 today is included in Section III in Appendix 1 of the White  
8 Paper. This material is relevant to Charge Questions 3, 4, 5,  
9 and 6.

10 In this presentation, I'll give a brief  
11 overview of the Tier I Exposure Assessment Methodology,  
12 including a description of the purpose of a Tier I assessment,  
13 as well as a discussion of where this method fits into the  
14 decision framework that was presented earlier by Keith  
15 Sappington.

16 As Christina Wendel discussed earlier, dietary  
17 exposure is a major exposure route of pesticides for honey bees.

18 So in order to estimate pesticide exposures through the diet,  
19 it's necessary to understand what honey bees eat and how much.

20 In this presentation I'll describe the  
21 proposed food consumption rates for worker larvae and for  
22 adults. I'll also discuss how conservative these consumption  
23 rates are when compared to other honey bees, as well as other  
24 species of bees.

25 I'll then move on to describe the proposed  
26 method for estimating pesticide exposure from contact that



1 results from foliar applications as well as the methods for  
2 estimating pesticide concentrations in pollen and nectar,  
3 following applications that are made via foliar spray, seed  
4 treatment and soil applications.

5 These concentrations in pollen and nectar are  
6 used in combination with the proposed food consumption rates for  
7 adults and for larval worker bees in order to estimate doses  
8 that are received by these two groups of bees. When I'm saying  
9 "bees" here, I mean honey bees.

10 Now I'll give a brief overview of the Tier I  
11 Exposure Assessment. Now, the goal of the Tier I Method is to  
12 generate reasonably conservative estimates of pesticide  
13 exposures to bees. This method is intended to estimate upper  
14 bound concentrations that are comparable to an upper 95th  
15 percentile exposure for honey bees.

16 When evaluating whether a method generates  
17 reasonably conservative estimates of exposure, estimates should  
18 preferably be higher and generally be within one or two orders  
19 of magnitude of available exposure data, empirical exposure  
20 data.

21 The Tier I Exposure Assessment Method is  
22 designed to allow risk assessors to confidently distinguish  
23 between those pesticides that do not pose a risk to honey bees  
24 and those that require further consideration. In this approach,  
25 we attempt to minimize Type II errors, which would involve  
26 concluding that there is no risk when there actually is a



1 potential for effects. And at the Tier I level, Type I errors  
2 are more acceptable because additional refinements can always be  
3 made at higher tiers of the risk assessment.

4 Now, this figure is a subset of the decision  
5 framework for foliar spray applications that was presented  
6 earlier today by Keith Sappington. And the focus of this  
7 particular slide is on the Tier I Exposure Assessment Method.

8 As depicted in Boxes 1 and 2 of this figure,  
9 once the use information for a pesticide product is determined,  
10 the risk assessor would determine whether or not there's a  
11 potential exposure for adults or for brood to the pesticide. In  
12 making this determination, the risk assessor would need to  
13 consider various factors, such as the potential overlap of the  
14 pesticide application and the presence of honey bees on the  
15 field, the attractiveness of the crop to honey bees or plants  
16 that may be present in areas adjacent to the field, such as  
17 dandelions.

18 If there is the potential for exposure, the  
19 risk assessor moves on to boxes 3 a, b, and c, which is the  
20 focus of this particular method. These involve estimation of  
21 exposures to adults and to larvae. These three boxes are the  
22 focus of the Tier I Exposure Assessment Methodology. For foliar  
23 spray applications, exposures are assessed separately for  
24 contact and for diet.

25 As depicted in boxes 4 a, b, and c, estimated  
26 exposure values should be matched to their respective toxicity



1 endpoints from contact and oral tests for adult honey bees and  
2 larvae to derive risk quotients.

3 Tier I toxicity endpoints are expressed on a  
4 dose basis in units of micrograms active ingredient, or AI, per  
5 bee. Therefore, all exposure values are expressed on a dose  
6 basis. Boxes 4 b and c indicate that both acute and chronic  
7 risk quotients are calculated in the Tier I risk assessment.  
8 Both acute and chronic exposure estimates are represented by the  
9 highest single days' dose.

10 Although a time-weighted estimated exposure  
11 value may be more representative of the exposure period in a  
12 chronic toxicity test, exposure occurring over a single day  
13 could potentially be sufficient to elicit effects. Therefore,  
14 in the Tier I approach, chronic exposure is conservatively  
15 represented by the highest single day estimated exposure.

16 This figure is a subset of the decision  
17 framework for seed and soil treatments, again, with a focus on  
18 the Tier I Exposure Assessment Method. I'm not going to go into  
19 detail on this figure because it's very similar to the one I  
20 just discussed for foliar applications. But the one notable  
21 difference here is that there's no contact exposure that's  
22 included in this decision tree. So the major exposure route  
23 incorporated into the Tier I Exposure Assessment Method for soil  
24 and seed treatments is through the diet.

25 Now I'll move on to discuss the honey bee food  
26 consumption analysis. And this is covered in detail in Appendix



1 1 of the White Paper. The basis of the honey bee diet is pollen  
2 and nectar. Honey bees will consume pollen and nectar directly  
3 or they'll eat bee bread and honey, which are forms of pollen  
4 and nectar that are processed for storage in a hive.

5 In addition, honey bees consume royal jelly  
6 and brood food, which are composed primarily of glandular  
7 secretions that are produced by nurse bees. The amount of food  
8 and specific consumption of a honey bees' diet varies by caste  
9 and by age. The diet of a worker bee changes over the course of  
10 its life to meet the energy requirements of its task which  
11 change with age.

12 For instance, nurse bees consume the most  
13 pollen, compared to other worker bees, so that they can produce  
14 jelly to feed larvae and the queen. Foraging bees consume the  
15 most honey and nectar in order to meet the energy demands of  
16 flying.

17 Because the proposed risk assessment method  
18 involves conducting separate risk assessments for brood and for  
19 adults at the Tier I level, separate food consumption rates for  
20 larvae and for adults are necessary to calculate the different  
21 pesticide exposures through the diet.

22 The proposed food consumption rates for the  
23 Tier I Exposure Assessment are 292 milligrams of food per day  
24 for adults and 120 milligrams of food per day for larvae. As I  
25 indicated earlier, the Tier I Exposure Assessment is intended to  
26 produce reasonably conservative estimates of exposure.



1                   Therefore, the proposed food consumption rates  
2 are based on portions of the adult and larval life stages where  
3 honey bees are expected to be consuming the greatest amounts of  
4 food, and would therefore be expected to receive the highest  
5 pesticide exposures through the diet, when compared to honey  
6 bees at different ages.

7                   So for adults, the nectar forager bees consume  
8 the most food and for larvae, it's individuals that are aged  
9 five days. There are several assumptions related to the  
10 proposed food consumption rates. In this approach, it is  
11 assumed that exposures through consumption of nectar and pollen  
12 are conservative representations of potential exposures through  
13 consumption of honey and bee bread.

14                   This approach is likely to be conservative  
15 because it assumes that pesticides contained in honey and bee  
16 bread do not degrade while these foods are stored in the hive.

17                   For honey bees that consume honey, it's  
18 assumed that the estimated pesticide exposures can be related  
19 back to the original concentration of the pesticide and nectar.  
20 This is accomplished by accounting for the amount of sugar  
21 that's consumed by bees and the amounts of sugar that are  
22 present in honey and in nectar. This is necessary since  
23 pesticide concentrations are estimated in nectar of treated  
24 crops.

25                   Using this approach, the pesticide dose  
26 received by a honey bee consuming nectar would be a equivalent



1 to a dose received by a honey bee consuming honey from the same  
2 source of nectar. It's also assumed that pollen and nectar  
3 consumption rates and resulting exposures are protective of  
4 exposures of honey bees, the pesticides through consumption of  
5 royal jelly and brood food. This is based on results from two  
6 studies that have observed concentrations of pesticides in royal  
7 jelly that were 100 times less than the food that was consumed  
8 by the nurse bees that produce that jelly.

9 Finally, for the Tier I Exposure Assessment  
10 Method, we're proposing to use the estimated pesticide  
11 concentrations in or on crop foliage as a surrogate for  
12 pesticide concentrations in nectar and in pollen.

13 So essentially, the pesticide concentration in  
14 nectar is treated in the Tier I method as being equivalent to  
15 the concentration in pollen. Therefore, for the Tier I Exposure  
16 Assessment Method, the proposed food consumption rates are based  
17 on consumption of pollen plus nectar. So I'll express that as  
18 total food consumption.

19 In order to indentify the group of adult  
20 worker bees that have the highest food consumption rates, pollen  
21 and nectar consumption information were identified in the  
22 scientific literature. Now, this table provides food  
23 consumption rates for different adult worker bees, as identified  
24 by their task.

25 The age range associated with each task is  
26 intended to be general and basically reflects the task that a



1 worker will carry out at different ages of her life, with  
2 in-hive tasks being carried out by younger bees and foraging  
3 tasks that occur outside of the hive being carried out by older  
4 worker bees.

5 Pollen consumption rates of worker honey bees  
6 are based on direct empirical measures published by Crailsheim,  
7 et al. in 1992. Nectar consumption rates are based on a  
8 commonly cited body of work published by Rortais, et al. in  
9 2005.

10 These authors estimated sugar consumption  
11 rates of different types of worker honey bees based on energy  
12 requirement that were associated with the bees' specific tasks.  
13 The sugar consumption rates published by Rortais, et al. are  
14 converted here to nectar equivalence by accounting for the  
15 average sugar content of nectar, which is 30 percent. As you  
16 can see by this table, pollen consumption rates vary among bees,  
17 but are at least one order of magnitude below nectar consumption  
18 rates of the same bees.

19 When we compare the total daily food  
20 consumption rates among the different worker bees, the nectar  
21 foraging bees had the highest food consumption rates. As you  
22 can see by this table, the majority of food consumption of this  
23 group of bees is represented by nectar, since the pollen  
24 consumption rate is four orders of magnitude lower.

25 Since the food consumption rate has a large  
26 influence on the final estimated dose received by honey bees and



1 for the nectar forager, this food consumption rate is  
2 represented by nectar consumption. We did a detailed review of  
3 the analysis conducted by Rortais, et al. for the nectar forager  
4 bee.

5 According to the method published by Rortais,  
6 et al., the nectar consumption rate of the nectar foraging bee,  
7 as estimated, using five variables, including the sugar required  
8 for flying, the amount of sugar that's required for flying. The  
9 number of foraging trips that are made in one day, the duration  
10 of a single foraging trip, the fraction of time spent flying  
11 during a trip, and the amount of sugar present in nectar.

12 One limitation to the approach is that there's  
13 a great deal of variability in these parameters. Also, the  
14 approach presented by Rortais, et al. did not account for the  
15 sugar requirements of bees while at rest.

16 Because there's so much variability within  
17 each of these parameters, we conducted a distributional analysis  
18 using a Monte Carlo simulation that simultaneously accounted for  
19 variability in all five parameters. We also accounted for the  
20 resting metabolic requirements of nectar foragers.

21 The results of the Monte Carlo simulation are  
22 depicted in this figure. The proposed food consumption rate for  
23 adult worker bees is 292 milligrams of food per day. This value  
24 is based on the median estimate of nectar consumption of nectar  
25 foraging bees in the simulation that was conducted.

26 The median value is selected for two primary



1 reasons. First, the median is considered most likely to  
2 represent food consumption of this group of bees. Second, use  
3 of the median avoids compounding conservative assumptions in the  
4 overall Tier I Exposure Method.

5 The proposed food consumption value already  
6 targets the group of bees with the highest food consumption  
7 rates compared to other adult bees. As I indicated earlier, the  
8 goal of the Tier I Exposure Assessment Method is to estimate  
9 reasonably conservative exposures to bees.

10 Now, the target of the reasonably conservative  
11 estimate is approximately the upper 95th percentile, and also  
12 I'll discuss later, the estimated pesticide concentrations in  
13 pollen and nectar represent an upper bound.

14 If an upper bound food consumption rate for  
15 nectar foragers was also proposed, the final dietary dose would  
16 most likely exceed the desired 95th percentile for worker bees  
17 due to a combination of a upper bound estimate of residues and  
18 food with an upper bound estimate of food consumption.

19 Next, I'd like to discuss the relative  
20 conservativeness of the proposed food consumption rate for adult  
21 bees when compared to other worker honey bees and drones, as  
22 well as other species of bees. This figure depicts the nectar  
23 and pollen consumption rates of different adult worker honey  
24 bees and drones.

25 As you can see by this figure, the worker bee  
26 that forages for nectar has the highest food consumption rate



1 among adult worker bees and drones, even when considering nectar  
2 and pollen consumption rates.

3 Nectar foraging bees are obviously summer  
4 bees. Winter bees that maintain the functions of the hive have  
5 different energy requirements. The nectar forager food  
6 consumption rate is higher than the food consumption rate of  
7 winter workers, indicating that it is also conservative for  
8 winter bees.

9 These data are presented in Appendix 1 of the  
10 White Paper. In some cases, ranges of food consumption rates  
11 were presented in this appendix. In this figure, the midpoints  
12 of those ranges are presented. This is worth noting here  
13 because the high-end estimate of nectar consumption of flying  
14 adult drones actually exceeds the proposed nectar consumption  
15 rate of nectar foraging workers; however, the low-end of the  
16 drone's nectar consumption rate is below the proposed value.

17 Therefore, the proposed food consumption rate  
18 for the nectar forager is still considered to be representative  
19 for drones. One limitation to this analysis is that data were  
20 not located to estimate the food consumption rates of adult  
21 queens. And this represents a data gap.

22 However, given that queens primarily consume  
23 royal jelly and that available data suggests that this food will  
24 contain at least 100 times lower pesticide concentrations,  
25 compared to the food that's consumed by nurse bees, the food  
26 consumption rate of queens would have to be at least 100 times



1 greater than the proposed food consumption rate for the nectar  
2 forager in order for the queen to receive a higher pesticide  
3 dose through the diet. Although the queen likely consumes more  
4 food than other honey bees, it seems unlikely that she would  
5 consume two orders of magnitude more food.

6 In summary, it appears that the proposed food  
7 consumption rate is conservative for other adult worker honey  
8 bees, as well as drones and queens. Before moving on, it's  
9 worth noting here that this talk is focused on the food  
10 consumption rates for the Tier I Exposure Assessment. In some  
11 cases, the risk assessor may choose to use different food  
12 consumption rates than the proposed value.

13 For instance, if chemical-specific pesticide  
14 concentration data are available for pollen and nectar, the risk  
15 assessor may want to use the pollen and nectar consumption rates  
16 separately. In addition, the food consumption rates that are  
17 depicted here for different worker bees could also be used for  
18 colony level modeling.

19 As I noted previously, honey bees also consume  
20 honey and that we're calculating food consumption rates on a  
21 nectar-equivalent basis. When we consider the relative food  
22 consumption rates among worker bees, when honey and pollen  
23 consumption is considered, the nectar forager is still the most  
24 conservative among adult bees.

25 We also compared the proposed food consumption  
26 rate for adult worker honey bees to available food consumption



1 rates for three other species of bees, including the bumblebee,  
2 the European mason bee and the alfalfa leaf-cutting bee.

3 These data suggest that the proposed food  
4 consumption rate for adult honey bee workers is similar to that  
5 of the adult bumblebee and is greater than that of the European  
6 mason bee and the alfalfa leaf-cutting bees.

7 This information suggests that the proposed  
8 food consumption rate for the Tier I Exposure Assessment for  
9 adult honey bees would be representative and somewhat protective  
10 for adults of these three non-Apis species. Please note that  
11 this information is discussed in Section 5.3 of the White Paper  
12 if you'd like any additional details.

13 As with adult worker bees, the intention of  
14 the food consumption rate for honey bee larvae is to focus on  
15 the age of larvae that consume the most food compared to other  
16 ages. Over the course of the five-day unkempt period, the diet  
17 of larvae changes. During the first three days they eat brood  
18 food and royal jelly. During the fourth and fifth days, they  
19 eat honey and pollen.

20 Worker larvae grow at an exponential rate over  
21 the course of their five-day developmental period. It's assumed  
22 that their food consumption rate doubles each day in order to  
23 meet the metabolic requirements of growth. The food consumption  
24 rates of larvae are based on information cited by Rortais, et  
25 al. that indicated that larvae consumed 120 milligrams of food  
26 over the course of the fourth and fifth days.



1           The pollen consumption rates are based on  
2       empirical measures of pollen contained in the guts of larvae.  
3       It is assumed that the remainder of the 120 milligrams of food  
4       consumed over days four and five is honey that is diluted with  
5       sugar -- with water, to 45 percent sugar. Based on the  
6       assumption that the food consumption rate of larvae doubles  
7       every day, the food consumption rates were calculated as  
8       depicted in this table.

9           The proposed food consumption rate of 120  
10       milligrams of food per day is based on the food consumption of  
11       the larvae at Day 5. This value represents consumption of 2.7  
12       milligrams of pollen per day and 117 milligrams of nectar per  
13       day. In this case, the amount of honey consumed is converted to  
14       nectar equivalence, assuming that the sugar content of nectar is  
15       30 percent and the sugar content of honey, which is diluted with  
16       water, is 45 percent.

17           Next, I'd like to discuss the relative  
18       conservativeness of the proposed food consumption rate for  
19       worker larvae when compared to other castes of honey bees and  
20       then to other species of bees. This figure depicts the total  
21       food consumption rates of worker larvae at different days of  
22       this life stage.

23           Please note that at on the X-axis, the caste  
24       of the larvae is provided along with the day of development in  
25       parentheses. The Y-axis represents food consumption rates where  
26       food is represented by royal jelly or brood food, honey, and



1 pollen.

2 This figure includes the food consumption  
3 rates for drone larvae at days 5 and 6. The food consumption  
4 rate for the five-day-old worker larvae appears to be consistent  
5 with that of the drone larvae. It should be noted here that the  
6 pollen consumption rates of drone larvae are unknown. In this  
7 figure, it's assumed that they are equal to the pollen  
8 consumption rates of workers at day 4 and 5.

9 Given that pollen consumption rates are  
10 generally much lower than honey consumption rates, this  
11 assumption is unlikely to impact the conclusion that the  
12 five-day worker larval consumption rate of food is consistent  
13 with that of drone larvae.

14 This figure includes the food consumption  
15 rates for queen larvae at days 1 through 5. These food  
16 consumption rates are calculated by assuming that queen larvae  
17 consumed 2.5 times the amount of food that's consumed by worker  
18 larvae at the same age. Therefore, the food consumption rate of  
19 queen larvae at day 5 is higher than that of the worker larvae  
20 at day 5.

21 When we convert the honey consumption rates of  
22 worker and drone larvae to a nectar equivalent basis, the  
23 five-day worker larvae value is still less than the five-day  
24 queen larval value for consumption of royal jelly. This is a  
25 concern because we don't want to have exposure values that pass  
26 screens, but the colony may be impacted due to elevated



1 pesticide doses received by the queen. This is a case where a  
2 Type II error could occur.

3 Although the queen larvae consume more food  
4 compared to worker larvae at day 5, the two types of larvae are  
5 consuming different types of food. As I noted earlier, we have  
6 data available from, two different studies, involving pesticides  
7 that indicated that pesticide concentrations and royal jelly are  
8 at least 100 times lower compared to the food fed to nurse bees.

9 On the next slide I will illustrate the impact of diet and  
10 relative concentrations on the different foods.

11 This figure depicts the relative doses  
12 received by larvae through consumption of royal jelly or brood  
13 food at a concentration of one microgram active ingredient per  
14 kilogram and pollen and nectar at a concentration that's 100  
15 times greater.

16 Although the consumption rate of queen larvae  
17 at day 5 is higher than worker larvae by a factor of 2.5, the  
18 overall dose received by the queen larvae is less than the  
19 worker larvae because the pesticide concentration in the queen  
20 larvae's food is so much lower.

21 In summary, the proposed food consumption rate  
22 for the Tier I Exposure Assessment for larval honey bees appears  
23 to be conservative compared to other castes of honey bees. We  
24 also compared the proposed food consumption rate for larval  
25 honey bees -- which is depicted in the top row of this table --  
26 to available food consumption rates for larvae of other species



1 of bees, including the bumblebee, the European mason bee and the  
2 alfalfa leaf-cutting bee.

3 These data suggest that the proposed food  
4 consumption rate for larval worker honey bees is representative  
5 or even protective compared to food consumption rates of these  
6 three species of bees.

7 To summarize, the food consumption rate  
8 analysis, the proposed method assumes that at the Tier I level,  
9 food consumption can be based on nectar and pollen. This  
10 assumes that honey bees are either eating these foods directly  
11 or that they're eating stored forms of these foods.

12 It is assumed that pesticide concentrations in  
13 pollen and nectar are equal. This also assumes that pesticide  
14 concentrations in brood food and royal jelly are less than those  
15 in nectar and pollen. The proposed food consumption rate for  
16 adult honey bees is 292 milligrams per day, based on the nectar  
17 foraging bee. The proposed larval food consumption rate of 120  
18 milligrams per day is based on five-day old larvae.

19 Compared to other castes of honey bees and  
20 three other species of bees, these values appear to be  
21 conservative. With Charge Question 5, EPA, PMRA and Cal DPR are  
22 seeking feedback from the SAP on the proposed food consumption  
23 rates for the Tier I Assessment, including consideration of the  
24 calculations behind these values, their assumptions, their  
25 strengths and limitations, and their conservativeness relative  
26 to other castes of honey bees.



1                   In Charge Question 3, we are interested in  
2 feedback from the SAP on the extent to which the honey bee can  
3 be a surrogate for assessing risk to other species of bees.  
4 We're interested in feedback on the extent to which the  
5 available food consumption rates indicate that dietary doses  
6 predicted for honey bees at the Tier I assessment level will be  
7 protective for other species of bees.

8                   Now I'll move on to discuss the proposed Tier  
9 I Exposure Assessment Methods for estimating pesticide doses  
10 received by honey bees through contact or dietary exposure. The  
11 proposed contact dose for foliar spray applications is based on  
12 the maximum dose on forager honey bees from a study published by  
13 Koch and Weisser in 1997.

14                  Dietary exposure is assessed for foliar spray,  
15 seed treatments and soil applications. All three application  
16 types have different methods for estimating pesticide  
17 concentrations in pollen and nectar which are converted to doses  
18 using the proposed food consumption rates I just discussed for  
19 adult and larval honey bees.

20                  In all three methods, estimated pesticide  
21 concentrations in or on plant foliage of treated crops is used  
22 as a surrogate for pesticide concentrations in nectar and  
23 pollen. For foliar spray applications, the proposed approach  
24 involves the use of the tall grass concentration from the T-REX  
25 model, as a surrogate for pesticide concentrations in nectar and  
26 pollen.



1                   For seed treatments, the proposed  
2                   concentration is one-milligram a.i. per kilogram in nectar and  
3                   pollen based on EPPO's default screening concentration. EPPO  
4                   stands for the European and Mediterranean Organization for Plant  
5                   Protection. For soil treatments, the proposed method is based  
6                   on a modification to the plant soil uptake model developed by  
7                   Briggs, et al., and published in 1982 and 1983.

8                   This model is designed to estimate pesticide  
9                   concentrations in plant shoots. When explaining each exposure  
10                  method, I'll follow a similar format. I'll start out with a  
11                  description of the proposed approach, followed by an  
12                  evaluation of the method using relevant empirical data. I'll  
13                  then discuss the assumptions and uncertainties associated with  
14                  the method and then its strengths.

15                 In determining which methods to consider for  
16                 the Tier I Exposure Assessment, we considered approaches that  
17                 have been peer reviewed with an emphasis on models that have  
18                 been vetted in a regulatory context. As I'll discuss, various  
19                 methods were evaluated by comparing estimated exposures to  
20                 available empirical data for honey bees, nectar and pollen to  
21                 determine whether the estimates could be considered reasonably  
22                 conservative.

23                 To compare the estimated exposures to the  
24                 empirical measurements, all exposure values were normalized to  
25                 an application rate of one point a.i. per acre. Empirical data  
26                 were available from the scientific literature as well as



1 unpublished registrant-submitted studies. In some cases, the  
2 identity of pesticides are disguised here and in the White Paper  
3 in order to maintain confidentiality of pesticides that are not  
4 yet registered.

5 The level of detail and amount of data  
6 provided in each study varied from study to study. The amounts  
7 of data that could be used to evaluate the methods also varied,  
8 which led to differences in the evaluation of the  
9 conservativeness of the methods.

10 For example, for foliar spray applications,  
11 the empirical data available to evaluate the proposed methods  
12 for estimating exposures through consumption of pollen and  
13 nectar were much larger than the data for estimating contact  
14 exposures.

15 For foliar spray applications, the proposed  
16 method for estimating exposures to adult forager bees through  
17 contact is to use the highest dose from a field study published  
18 by Koch and Weisser in 1997. In this approach, the contact dose  
19 is 2.7 micrograms a.i. per bee, per one pound a.i. per acre. In  
20 applying this method, the actual dose is calculated by  
21 multiplying the single highest application rate in pounds a.i.  
22 per acre by 2.7.

23 The study conducted by Koch and Weisser  
24 involved measuring the concentration of a tracer on honey bees  
25 soon after foraging in the treated area. Honey bees were  
26 collected, following applications to phacelia fields and apple



1 orchards that were located in Germany.

2 The study involved multiple field trials that  
3 were conducted over the course of several years. During this  
4 study, tracer residues were quantified on over 6,000 honey bees.

5 The proposed dose represents the highest value that was  
6 measured during this study. We chose the highest dose rather  
7 than the upper bound due to some uncertainties associated with  
8 this data set, in particular, because the study only includes  
9 two crops. In our application of this methods, we want to be  
10 able to represent multiple crops.

11 As indicated by comparing the doses observed  
12 in the trials on the apple and phacelia fields, the distribution  
13 of doses may vary from crop to crop. This figure depicts the  
14 frequency distribution of residues measured on honey bees  
15 foraging on phacelia fields over the course of the five trials  
16 that were conducted by Koch and Weisser.

17 As you can see from this figure, 43 percent of  
18 the honey bees analyzed had residues that were in order of  
19 magnitude below the proposed dose of 2.7 micrograms a.i. per  
20 bee, normalized to one pound a.i. per acre.

21 This figure depicts the frequency distribution  
22 of residues that were measured on honey bees foraging on apple  
23 orchards over the course of the nine trials that were conducted  
24 during this study. As you can see from this figure, 71 percent  
25 of all of the honey bees that were analyzed had residues that  
26 were in order of magnitude below the proposed dose of 2.7.



1           These results, along with those from the  
2 phacelia trial suggest that the proposed dose is conservative  
3 when considering all of the results from this study. In  
4 developing the proposed method for assessing contact exposures  
5 to honey bees following foliar application, we also considered  
6 the utility of the upper bound arthropod exposure value that is  
7 in the current version of the T-REX model.

8           This dose is 12 microgram a.i. per bee, per  
9 one pound a.i. per acre, which is a factor of five higher than  
10 the proposed contact dose from the Koch and Weisser dataset.  
11 This dose, the T-REX dose is calculated by multiplying the upper  
12 bound concentration of 97 milligrams active ingredient per  
13 kilogram of arthropods by the average body weight of an adult  
14 worker bee, which is .128 grams.

15           The upper bound pesticide concentration for  
16 arthropods used in the T-REX model represents a 95th percentile  
17 concentration on arthropods, based on empirical data from the  
18 scientific literature and registrant-submitted studies. These  
19 studies measured pesticide residues on various arthropods,  
20 including moth and beetle larvae, crickets, grasshopper, beetles  
21 and unidentified arthropods that were captured in treated areas.

22       This dataset does not include residue data for honey bees.

23           The use of the arthropod concentration relies  
24 upon measured residues of pesticides in and on arthropods that  
25 were located on treated fields at the time that the field was  
26 sprayed. It is assumed that the residues are predominately



1 based on direct spray; however, they may also incorporate some  
2 contact of arthropods with pesticide residues on treated  
3 foliage, as well as consumption.

4 We chose not to propose the use of the T-REX  
5 arthropod dose because it does not include residues from honey  
6 bees. Since the focus of the Tier I risk assessment method is  
7 on honey bees, the proposed dose was based on the Koch and  
8 Weisser data.

9 In cases where it is necessary to estimate  
10 exposures to other species of bees, the T-REX approach may be  
11 more representative of residues resulting from contact exposure.  
12

13 A review of the scientific literature failed  
14 to identify maximum doses following contact exposures of honey  
15 bees to pesticides resulting from foliar spray applications.  
16 Two studies were identified that contained mean residues of  
17 pesticides on bees.

18 The figure depicted on this slide compares the  
19 mean doses reported in those studies to the mean doses from the  
20 Koch and Weisser study on phacelia and apple orchards, as well  
21 as the T-REX arthropod dose. The mean doses all appear to be  
22 similar, generally being within an order of magnitude of the  
23 Koch and Weisser data.

24 The method evaluation for the proposed contact  
25 exposure dose is uncertain because there are a limited number of  
26 studies available to evaluate the approach. Of the two studies



1 that are available, only mean measured concentrations on honey  
2 bees are reported.

3 In addition, because there are only two  
4 studies available for evaluation purposes, it's unclear how  
5 representative these data are related to other locations, crops,  
6 and field conditions.

7 It's unknown whether the data from these two  
8 studies are representative of high-end exposures that other  
9 honey bees may receive under different conditions. Despite  
10 these limitations, the Koch and Weisser maximum dose is  
11 empirically based and appears to be consistent with the data  
12 from the other two studies.

13 Also, the proposed dose appears to be  
14 conservative when we consider the overall distributions of  
15 residues that were detected by the authors over the course of  
16 the study, namely that the majority of the contact doses  
17 measured on the foraging honey bees were in order of magnitude  
18 lower than the highest dose.

19 The Koch and Weisser study appears to have a  
20 robust study design. The use of the tracer facilitated  
21 measurements of the full distribution of concentrations of the  
22 chemical on honey bees. This would not necessarily be the case  
23 for a fast-acting insecticide, where honey bees with lethal  
24 doses would not return to the hive and would not be measured.

25 In addition, this dose is based on a large  
26 end. Each trial quantified doses on hundreds of honey bees for



1 a total of more than 6,000 measurements on individual bees over  
2 the course of several years. In addition, the proposed contact  
3 dose is consistent with other approaches, including the T-REX  
4 arthropod dose as well as the Atkins Method.

5 For dietary exposure, the proposal includes  
6 the use of an upper bound residue value for foliage that is used  
7 in the T-REX model. T-REX is already used by the EPA for  
8 ecological risk assessments of pesticides for establishing  
9 screening level exposures to terrestrial vertebrates.

10 T-REX includes pesticide concentrations on  
11 different plants or plant parts, including short and tall grass,  
12 broadleaf plants, fruit, pods and seeds. The upper bound and  
13 mean concentrations used in T-REX are provided in this table.  
14 These concentrations are based on a review of pesticide residues  
15 on plants that was conducted by Kenega and then later updated by  
16 Fletcher, et al.

17 Ideally, the proposed approach would be to  
18 base the upper bound concentrations on measured values from  
19 pollen and nectar; however, there was insufficient amount of  
20 empirical data to confidently estimate the upper bound.  
21 Therefore, it's necessary to use a surrogate.

22 This figure depicts the method evaluation for  
23 the T-REX pesticide concentrations for short grass, broadleaf  
24 plants, tall grass and fruit pods and seeds. The estimated  
25 concentrations are depicted in this figure with red dashed  
26 lines. And you can see to the right of the dash which



1 particular residue from T-REX is represented by the specific  
2 line.

3 The empirical data that are relevant to this  
4 method are depicted by blue columns. I'll use this format in  
5 other figures throughout the rest of this presentation.

6 The empirical data that are depicted here for  
7 nectar includes mean concentrations. Some maximum values are  
8 also available. Most often, only mean concentrations were  
9 reported in the literature.

10 As you can see by this figure, the mean  
11 concentrations for short grass, broadleaf plants and tall grass  
12 are all greater than the mean concentrations available for  
13 nectar. The highest empirical measurement from these studies  
14 was 13.6 milligram per kilogram nectar, normalized to one pound  
15 a.i. per acre. This value is on the same order of magnitude as  
16 the mean concentrations for tall grass, short grass, and  
17 broadleaf plants.

18 Four of the ten mean concentration of the  
19 empirical dataset exceeded the mean T-REX residue value for  
20 fruit, seeds, and pods. All of the empirical concentrations  
21 from the maximum dataset were below the T-REX upper bound  
22 concentrations for all four categories.

23 This figure depicts the mean empirical  
24 concentration data for pollen, compared to the mean T-REX  
25 concentrations. The mean concentration for short grass is  
26 greater than the mean concentrations available for all of the



dataset for pollen. The mean concentrations for broadleaf plants and tall grass exceed all but one of the nine mean empirical concentrations for pollen.

Five mean pollen concentrations exceed the mean T-REX concentration for fruit, seeds, and pods. All of the maximum empirical concentrations for pollen are below the upper bound concentrations for short grass, broadleaf plants and tall grass. Seven of the 14 maximum concentrations exceed the upper bound concentration for fruit pods and seeds.

So when considering all of the empirical data that are available for nectar and pollen, the T-REX concentrations for short grass, broadleaf plants and tall grass are consistently above the empirical concentrations. Only one value out of more than 40 exceeds the tall grass and broadleaf plant concentrations.

The tall grass upper bound concentration is proposed as a surrogate because of the three possible surrogates that appear to be conservative for nectar and pollen. Tall grass is the closest to the empirical data.

For the Tier I Exposure Assessment, the upper bound concentration of 110 milligram a.i. per kilogram, normalized to one pound a.i. per acre, would then be converted to a dietary dose received by adult and worker bees using pollen and nectar consumption rates for these two life stages. Therefore, the dietary-based dose for larvae is 13 microgram a.i. per bee, per one pound a.i. per acre, and the value for



adults is 32.

Estimated doses can be scaled to an application rate by multiplying by the maximum single application rate. And in practice, the concentrations generated by T-REX also account for multiple applications and dissipation of the chemical in between applications.

In terms of assumptions and uncertainties associated with this method, I've already referenced the most significant one, which involves the assumption that the upper bound concentration for tall grass will be an appropriate surrogate for pesticide concentrations in pollen and nectar of flowers that were directly sprayed with the pesticide.

Comparison of empirical data to the tall grass mean and upper bound concentrations suggests that this value is reasonably conservative. As empirical measures of pesticide concentrations on pollen and nectar become available, we may be able to refine the Tier I Exposure Assessment Method using pollen or nectar-specific data rather than using plant foliage as a surrogate.

Also, in using the T-REX concentration, it's assumed that direct spray onto foliage represents a conservative estimate of concentrations in pollen and nectar for both systemic and non-systemic pesticides.

Although systemic transport of pesticides to pollen and nectar are expected, concentrations in nectar and pollen that occur due to systemic transport are not expected to



1 exceed the concentrations from direct spray. This is because  
2 systemic transport takes time and not all of the amounts of the  
3 pesticide that is applied to the plant will move the pollen and  
4 nectar due to dissipation of the pesticide and transport to  
5 other portions of the plant.

6 The proposed Tier I Exposure Method for seed  
7 treatments is based on the EPPO 2010 screening concentration of  
8 one milligram a.i. per kilogram and pollen and nectar of plants  
9 to which seed treatments were made. EPPO screening  
10 concentration of one milligram a.i. per kilogram is based on the  
11 maximum value from data compiled by Alix, et al. in 2009,  
12 including pesticide residues measured in different plant parts  
13 following applications to soil or seed treatments.

14 Plant parts included in this analysis were  
15 described as leaves, fruit, green parts -- in parenthesis --  
16 whole plants and grains. This approach may be used for all  
17 pesticides that are applied via seed treatment, by assuming that  
18 the upper bound concentration for pollen and nectar is one  
19 milligram a.i. per kilogram with no need for adjustment, based  
20 on application rate or chemical properties.

21 This concentration can then be multiplied by  
22 the food consumption rates for adult and larval worker bees to  
23 determine the upper bound doses potentially received by honey  
24 bees. For adults, this dose is .29 microgram a.i. per bee. And  
25 for larvae, the dose is .12.

26 EPPO screening concentration was evaluated



1 using empirical data describing pesticide concentration in  
2 pollen and nectar of crops whose seeds were treated with the  
3 pesticide. The highest concentration measured in pollen was  
4 .036 milligram a.i. per kilogram, and again, no adjustment was  
5 made for application rate here. This value is a factor of 28  
6 lower than the EPPO screening value.

7 Considerably less empirical data are available  
8 to compare the EPPO one milligram a.i. per kilogram screen to  
9 nectar concentrations. The screening concentration is three  
10 orders of magnitude above the highest concentration for nectar,  
11 which is .003 milligram a.i. per kilogram.

12 In the Tier I approach, it's assumed that all  
13 pesticides that are applied to seeds are systemic, and therefore  
14 can be transported into pollen and nectar that may be consumed  
15 by honey bees. If this is not the case for a specific  
16 pesticide, registrants can submit data to refute the assumption  
17 of systemicity.

18 The proposed use of the EPPO screening level  
19 exposure concentration of one milligram a.i. per kilogram for  
20 pollen and nectar of seed treated crops does not account for  
21 differences in application rate where the mass of the pesticide  
22 that's applied to a seed.

23 There is uncertainty in not accounting for the  
24 mass of the chemical applied since it's expected that the  
25 magnitude of the pesticide concentration of the plant will be  
26 influenced by the magnitude of the application. However, the



1 evaluation of this screen indicates that it is protected for  
2 rates that are currently being used for seed treatments which  
3 are approximately one milligram of active ingredient per seed.

4 Even if seed treatments would exceed one  
5 milligram a.i. per seed, it seems unlikely that pesticide  
6 concentrations and nectar and pollen would exceed the EPPO  
7 screening value since it is a factor of 28 greater than the  
8 highest residue that's measured in pollen and nectar.

9 In addition, pesticide mass applied to seeds  
10 is not expected to be that much greater than one milligram a.i.  
11 per seed, since this value is already similar to the overall  
12 weight of the seed itself.

13 Another uncertainty associated with this  
14 approach is that it does not account for physical chemical  
15 properties of a pesticide that may influence its potential  
16 systemic transport throughout the plant and into pollen and  
17 nectar. For example, a chemical with low mobility may be  
18 expected to have lower concentrations and nectar when compared  
19 to a chemical with higher mobility.

20 Despite these limitations, this approach  
21 appears to generate reasonably conservative estimates of  
22 pesticide concentrations in the pollen of seed treated crops.  
23 The one milligram a.i. per kilogram screening value is much  
24 greater than the data that are available for nectar; however,  
25 there are only a few empirical concentrations, and so it may not  
26 be appropriate to conclude at this time that this method is



unreasonably conservative for nectar.

For soil treatments, the proposed method involves the use of the Briggs soil plant uptake model. This model predicts the concentration of a chemical in the shoot of a plant, using the  $K_{ow}$  that chemical and its concentration in the water that is in contact with the plant's roots.

This approach is based on observations from studies conducted by Briggs et al. that involved uptake of 18 different pesticides into the shoots of barley plants. These pesticides included carbamate insecticides and phenylurea herbicides with log  $K_{ow}$  that ranged from  $-0.57$  to  $4.6$ .

Note that one of the major components of the Briggs equation is the transpiration stream concentration factor or TSCF, which is a ratio of the concentration of the chemical in the xylem of the plant to the concentration of the aqueous medium that's in contact with the roots.

Median TSCF values are calculated in the Briggs' published model, using the  $K_{ow}$  of the chemical.

Suggestions to modify the Briggs approach were made by Ryan, et al. in 1988. This involved adding an equilibrium partitioning component based on the chemicals  $K_{oc}$  that allows for estimation of pesticide concentrations in plants, using concentrations in soil rather than in aqueous medium, as was used by Briggs.

Note that if  $K_{oc}$  is not available or appropriate for a chemical, the  $K_d$  can be used as an alternative



1 to the Koc and Foc component of this equation. The soil  
2 properties that are required by this equation can be easily  
3 obtained from EFED's standard scenarios that are used for the  
4 prism model.

5 The TSCF calculation provided by Briggs  
6 generates a median estimate of the TSCF in resulting  
7 concentration in plant shoots. As I noted at the beginning of  
8 this talk, the goal of the Tier I Exposure Assessment is to  
9 derive exposure estimates that represent an upper bound. To  
10 accomplish this, we recalculated TSCF values to represent the  
11 95th percentile values and they're depicted in this figure by  
12 the red line.

13 These TSCF values are provided in Appendix 5  
14 of the White Paper and can be selected using the Kow of a  
15 chemical. Empirical data for pollen were available for four  
16 pesticides applied to pumpkin, squash, tomato or melon.  
17 Comparison of estimated concentrations to these measured data  
18 indicates that model predictions are generally conservative  
19 compared to empirical data. There are three empirical  
20 concentrations that are higher than the estimated values, but  
21 they are still within an order of magnitude.

22 It should be noted here that some  
23 discrepancies between the estimated and empirical data may  
24 originate from several sources. For instance, none of the  
25 empirical data were from barley, which is the basis for the  
26 Briggs model. And the soil properties that are used in the



1 Briggs model were not matched to the studies that were used to  
2 generate the empirical data.

3 In addition, the empirical data represents a  
4 combination of parent and degradates, whereas the estimated  
5 concentrations are based on parent only. When comparing  
6 empirical concentrations for nectar to the estimated  
7 concentrations, the results are similar to those of the pollen  
8 analysis. Empirical concentrations for nectar were available  
9 for five pesticides that were applied to fuchsia, nasturtium,  
10 pumpkin, squash, cotton, and melon.

11 Comparison of estimated concentrations to  
12 empirical data indicates that the model predictions are  
13 generally conservative compared to the empirical data. There  
14 are two empirical values that are higher than the estimated  
15 values, but they are still within an order of magnitude. As I  
16 noted previously, the discrepancies between the estimated  
17 empirical values may be attributed to differences in soil  
18 properties and systemic transport of the crops that are  
19 different than barley.

20 In addition, in the case of imidacloprid, for  
21 which the estimated value is below one of the eight empirical  
22 concentrations, the estimated value is based on the parent  
23 alone, whereas the empirical values represent parent plus  
24 degradates.

25 In developing the proposed method for  
26 estimating pesticide concentrations in nectar and pollen of



1 crops that receive soil treatments of pesticides, we also  
2 considered using the EPP0 one milligram a.i. per kilogram  
3 screening concentration that's proposed for use with seed  
4 treatments. When the one milligram a.i. per kilogram default is  
5 compared to maximum concentrations from available studies where  
6 pesticides were applied to soil, all empirical concentrations  
7 for pollen are below the one milligram a.i. per kilogram value.

8 Note that this comparison did not involve  
9 adjustment of the concentrations for application rate. There is  
10 one concentration for nectar that exceeds the screen by a factor  
11 of almost five; however this concentration was from a study  
12 where the application rate of dimethoate was equivalent to 17  
13 pounds a.i. per acre, which is much higher than currently  
14 registered application rates for this chemical and most other  
15 insecticides in North America.

16 With the exception of the dimethoate data, all  
17 the measured concentration of pesticides in pollen and nectar of  
18 plants receiving soil applications are two orders of magnitude  
19 below the one milligram a.i. per kilogram value. Based on this  
20 information, it appears that the one milligram a.i. per kilogram  
21 default concentration is a reasonably conservative estimate for  
22 soil applications. However, for pesticides that have high  
23 applications, this method may under-predict the exposure.

24 In addition, this approach does not account  
25 for the physical chemical properties of a pesticide that may  
26 influence its potential systemic transport throughout the plant



1 and into pollen and nectar. For example, a non-ionic organic  
2 chemical with low mobility may be expected to have lower  
3 concentrations in nectar when compared to a chemical with higher  
4 mobility.

5 There are several notable limitations to using  
6 the modified Briggs approach. As with the use of the T-REX tall  
7 grass upper bound concentration for foliar spray applications,  
8 this approach assumes that estimated concentrations of a plant  
9 pesticide and plant foliage is an appropriate surrogate for  
10 concentrations in pollen and in nectar.

11 Comparison of empirical data to the estimated  
12 concentrations suggests that the Briggs approach is reasonably  
13 conservative, relative to concentrations that have been measured  
14 in pollen and nectar. In the Tier I approach, it's assumed that  
15 all chemicals may be systemically transported. This assumption  
16 may be limited based on a chemical's log Kow, -for example, for  
17 chemicals that have log Kow values above five, they may not be  
18 systemically transported.

19 But whether a chemical is transported,  
20 systemically in plants, could potentially be confirmed using  
21 empirical data that is submitted to EPA; for example, plant  
22 metabolism studies. However, it would be up to pesticide  
23 registrants to submit sufficient data to demonstrate that a  
24 pesticide is not systemic.

25 Another limitation is that this methodology is  
26 based on empirical data from only one type of plant. Uptake may



1 be different for different types of plants. However, the  
2 species that was used by Briggs, which is barley, is  
3 representative of a crop species that is likely to be grown on  
4 soils where pesticides are applied.

5 Also, the dataset used to establish the Briggs  
6 equation is based on a limited number of chemicals that  
7 represent only two classes of pesticides. Despite this  
8 limitation, the pesticides that were used in this approach are  
9 systemic and cover a wide range of log Kow values, leading to an  
10 approach that is applicable to a wide range of pesticides with  
11 the potential to be systemically transported by plants.  
12 However, the approach may have limited utility for ionic  
13 chemicals whose transport may not be predicted while using Kow  
14 and Koc.

15 The final limitation I'll discuss is that the  
16 proposed approach is based on passive transport of chemicals  
17 into the xylem.

18 This approach does not directly estimate  
19 pesticide concentrations in plants that are the result of phloem  
20 transport. One possible assumption that could be made in  
21 applying this model for estimating pesticide concentrations in  
22 plants would be to assume that chemical transport via phloem, is  
23 equivalent to that of xylem.

24 The alternative approach would be to use  
25 different models for neutral and ionic compounds such as those  
26 described by Briggs, et al. 1987; Kleier 1988; and Trapp, et al.



1 1995. Those citations are provided in the White Paper.

2           Despite the limitations discussed here, both  
3 the Briggs model with the modifications by Ryan, et al. and  
4 those proposed by EPA in the White Paper, as well as the EPPO  
5 one milligram a.i. per kilogram screening value, both appear to  
6 be useful tools for generating reasonably conservative estimates  
7 of pesticide concentration in plants after systemic transport  
8 from the soil. These approaches are consistent with the  
9 screening level assessment because they're both simple and  
10 efficient and are empirically based on data for pesticides.

11           The Briggs model had the advantage that it  
12 relies upon some basic physical chemical properties of  
13 pesticides and also accounts for the mass that's applied to the  
14 soil. Therefore, the Briggs model is the one that's proposed  
15 for estimating pesticide concentrations in pollen and nectar,  
16 following a soil treatment.

17           This table summarizes the proposed Tier I  
18 Exposure Methods. The values provided in the right column  
19 represent the resulting doses for each method, based on an  
20 application of one pound a.i. per acre. For foliar  
21 applications, the proposed Tier I Exposure Assessment involves  
22 quantification of pesticide doses to adult honey bees through  
23 contact exposure. This method is based on the maximum dose of  
24 2.7 microgram a.i. per bee, per one pound a.i. per acre that was  
25 measured on honey bees as recorded by Koch and Weisser.

26           For foliar spray applications, seed treatments



1 and soil treatments that proposed Tier I Exposure Assessment,  
2 quantifies pesticide doses received by adults and larvae through  
3 the diet. These methods involve translating estimated  
4 concentrations in pollen and nectar into doses by multiplying  
5 the concentrations by the food consumption rates of adults and  
6 larvae. For adults, the proposed food consumption rate is 292  
7 milligrams of food per day. And for larvae, that value is 120.

8  
9 For foliar spray applications, the proposed  
10 method involves using the T-REX tall grass upper bound  
11 concentration of the surrogate for pesticide concentrations in  
12 pollen and nectar. The resulting doses of 32 micrograms a.i.  
13 per bee and 13 micrograms a.i. per bee for adults and larvae can  
14 be adjusted to the use of the pesticide by accounting for the  
15 application rate of the chemical.

16 These doses are the highest among the proposed  
17 methods, indicating that if the application rate is held  
18 constant, then foliar spray applications would represent the  
19 highest potential exposures to honey bees at the Tier I Exposure  
20 Assessment level.

21 For seed and soil treatments of pesticides,  
22 it's assumed that the pesticides are systemically transported to  
23 pollen and nectar. For seed treatments, the proposed method is  
24 to use the EPPO screening value of one milligram a.i. per  
25 kilogram to represent pesticide concentrations in nectar and  
26 pollen of treated crops. No adjustment s made for application



1 rate or mass of chemical applied to the seed. Therefore, for  
2 all uses and chemicals, the doses would be .29 and .12 microgram  
3 a.i. per bee for adults and larvae respectively.

4 For soil treatments, the proposed method for  
5 estimating pesticide concentrations in nectar and pollen  
6 involves the use of the modified Briggs soil plant uptake model.

7 This model relies on a pesticide's application rate, Kow and  
8 Koc to estimate the concentration in plant shoots which is a  
9 surrogate for pollen and nectar.

10 As an illustration of the relative doses  
11 generated by this approach, the values depicted in this table  
12 are for dimethoate, which is a mobile chemical with a low Kow.  
13 In evaluating the proposed methods using empirical data, the  
14 proposed methods appear to be reasonably conservative.

15 As additional information becomes available,  
16 we may reevaluate the proposed methods, for example, as targeted  
17 monitoring data become available where pesticides are measured  
18 in pollen and in nectar as part of Tier II studies, it may be  
19 possible in the future to replace the use of pesticide  
20 concentrations on plant foliage as surrogates with empirical  
21 measures of pesticides and pollen and in nectar.

22 The EPA is interested in SAP feedback on the  
23 proposed Tier I Exposure Assessment Method that I discussed in  
24 this presentation and is presented in detail in Section 3 of the  
25 White Paper. For contact exposures resulting from foliar  
26 applications, which is the focus of Question 4, EPA, PMRA and



1 Cal DPR are interested in SAP comments on the strengths and  
2 limitations of the proposed approach that is based on the  
3 maximum of the Koch and Weisser data.

4 We're also interested in SAP feedback in the  
5 potential utility of the T-REX arthropod dose in assessing  
6 contact exposure to non-Apis bees as well as the honey bee. The  
7 focus of Charge Question 6 is on the proposed methods, meaning  
8 pesticide concentration in pollen and nectar, which are relevant  
9 to assessing dietary exposures.

10 We're requesting SAP feedback to the extent to  
11 which the T-REX tall grass upper bound concentration the EPPO  
12 one milligram a.i. per kilogram screen and the modified Briggs  
13 model can represent pesticide concentrations in pollen and  
14 nectar for foliar spray, seed treatment and soil applications  
15 respectively.

16 We're also interested in any other methods or  
17 data that may represent viable alternatives to these proposed  
18 methods or may be used to improve the proposed methods, perhaps,  
19 in the form of reducing assumptions or uncertainties.

20 If I have time, I can take questions.

21 **DR. DANIEL SCHLENK:** Thanks. Dr.  
22 Fefferman.

23 **DR. NINA FEFFERMAN:** Okay. Thank you. That  
24 was really interesting. I'm wondering, a lot of the estimates  
25 that I saw for how you were using characterizations of larval  
26 and adult uptake for consumption rates were for normal colony



1 processes. And I'm wondering if so, how you guys are dealing  
2 with non-normal but lifecycle dependent colony processes.

3 So gorging, having to leave the hive very  
4 quickly or swarming times when both the energy needs of an  
5 individual may be different and also the consumption rate may  
6 skyrocket for non-energetic process needs.

7 **MS. KRISTINA GARBER:** The proposed method is  
8 based on kind of the baseline functioning of the colony. It  
9 doesn't account for those scenarios like swarming.

10 **DR. NINA FEFFERMAN:** Can we stick something in  
11 there about that? Because if they accidentally poison  
12 themselves once a year that might be really bad.

13 **MS. KRISTINA GARBER:** That's part of why we're  
14 here. So if you have any advice on how we could potentially  
15 modify that and some data that you could point to or good  
16 sources that could provide another way to quantify the  
17 consumption rates, that would be appreciated.

18 **DR. NINA FEFFERMAN:** Sure. Thanks.

19 **DR. DANIEL SCHLENK:** Okay. Dr. Berenbaum.

20 **DR. MAY BERENBAUM:** So estimating the contact  
21 exposure doses based on models that derive from empirical  
22 studies that in some cases are 15 to 30 years old, is there any  
23 indication or is it a consideration that detection methods have  
24 changed so that the ability to detect residues is also a  
25 variable in this process?

26 **MS. KRISTINA GARBER:** Just for clarification,



1 are you talking about the Briggs model as being --

2 **DR. MAY BERNEBAUM:** That's one that was  
3 referenced to Atkins '82 [sic] in there. There are a couple of  
4 studies, Koch and Weisser from '97, I think. So these are older  
5 technologies. Is there some consideration of how chemical and  
6 analytical techniques change so that detection is improving?

7 **MS. KRISTINA GARBER:** I think that at this  
8 point, when we were looking at these studies, I think that the  
9 detection rates were sufficient to confidently estimate the  
10 values that were being detected.

11 If you have any concerns about the specific  
12 detection rates, some of the studies would definitely account  
13 for that. If that's an uncertainty in some of the methods, then  
14 certainly we could look at newer studies that may account for  
15 that uncertainty.

16 **DR. DANIEL SCHLENK:** Okay. Dr. James.

17 **DR. ROSALIND JAMES:** Your doses are really  
18 dose per day, right?

19 **MS. KRISTINA GARBER:** Yes.

20 **DR. ROSALIND JAMES:** I work with solitary bees  
21 and they make a pollen provision and it's really easy to  
22 calculate the total dose that larvae will have over their entire  
23 development period. Honey bees also only feed for five days.  
24 One thing that I was thinking about when you had the drone  
25 consumption rates and they're eating less than the workers as  
26 larvae and yet they're bigger, so you know they must be eating



1 more because they have to get bigger somehow. If the  
2 measurements are accurate, it must be because they eat longer,  
3 they're larvae for longer, and similarly with queens.

4 So they could potentially be exposed to higher  
5 amounts of pesticide as larvae. But if you're only looking at  
6 dose per day you would miss that, right?

7 **MS. KRISTINA GARBER:** In developing this  
8 model, we did go back and forth between the idea of the duration  
9 of the exposure we were trying to represent. I think that what  
10 you're saying is a valid point, you know, that the duration of  
11 the EEC or the exposure that we're looking at, it is relevant.  
12 And part of why we settled on day was that it could be matched  
13 to the acute toxicity test when we're only exposing the bees for  
14 a shorter period of time.

15 **DR. DANIEL SCHLENK:** Dr. Tarpy.

16 **DR. DAVID TARPY:** I have a couple of  
17 questions, actually. One is largely out of my own ignorance,  
18 not being a toxicologist. But knowing that that there is  
19 variation, sometimes substantial variation, among workers, not  
20 to mention, huge variation with different castes, and even  
21 larger variation among different pollinators of which honey bees  
22 are supposed to be a proxy in this means; in the in vitro  
23 studies, why is the denominator a per bee, per day basis rather  
24 than say, milligram per individual?

25 **MS. KRISTINA GARBER:** By "denominator" you  
26 mean the toxicity endpoint?



1                   **DR. DAVID TARPY:** Yeah. All of the  
2 recommendations here on the milligrams of active ingredient per  
3 bee. Why is it on a per individual basis rather than something  
4 more standardized that can translate across different life forms  
5 and different types of pollinators?

6                   **MS. KRISTINA GARBER:** I think that part of  
7 that is the historical basis of how honey bee toxicity tests are  
8 conducted where they tend to be on a dose per individual bee.  
9 That's how the residues are quantified.

10                  **DR. DAVID TARPY:** Okay. So it's just based on  
11 practice and historical purposes, but there is variation there  
12 but it's not captured in these types of bioassays.

13                  **MS. KRISTINA GARBER:** Just to clarify, by  
14 "variation," do you mean individual variability?

15                  **DR. DAVID TARPY:** Yeah. Bees weigh different  
16 from each other.

17                  **MS. KRISTINA GARBER:** Right.

18                  **DR. DAVID TARPY:** And I would presume, and  
19 again, correct me if I'm wrong, but that toxicity would be a  
20 function of weight, of body weight to some capacity, assuming  
21 that's even linear. It might even be nonlinear effects, right.

22  
23                  I just wanted to ask that question, not  
24 knowing what the answer was. So you seem to have answered that.

25  
26                  **MS. KRISTINA GARBER:** Okay. Presumably, you



1 would catch some of the variability in the confidence interval  
2 that would surround your LD50. That would be generated from  
3 that. So you'd probably get -- if there is a relationship  
4 between weight and toxicity, then that would be translated in  
5 the 95th percentile confidence around your LD50.

6 **DR. DAVID TARPY:** Per individual.

7 **MS. KRISTINA GARBER:** Correct.

8 **DR. DAVID TARPY:** Okay. The second question  
9 is combining the total food intake for the different adult  
10 caste, behavioral caste, especially with the pollen versus  
11 nectar, is there consideration in doing these bioassays for  
12 lipophilic versus hydrophilic compounds in those different  
13 phases that in some cases, combining those two things in total  
14 food consumption per day might not be captured for particular  
15 compounds that are being screened.

16 **MS. KRISTINA GARBER:** So I have to confess,  
17 you're asking a lot of toxicity questions that are related to a  
18 talk that Joe DeCant will be giving.

19 **DR. DAVID TARPY:** Okay.

20 **MS. KRISTINA GARBER:** If I may, would you mind  
21 deferring your question to that time? Maybe that will be  
22 covered.

23 **DR. DAVID TARPY:** Mm-hmm. No problem.

24 **MS. KRISTINA GARBER:** Thank you.

25 **DR. DANIEL SCHLENK:** Dr. Potter.

26 **DR. THOMAS POTTER:** I had a question about any



1 consideration that may have been made regarding physical  
2 chemical properties of the chemicals and the potential  
3 concentrations in nectar, given its importance, in terms of an  
4 exposure pathway. Something, for example, is the aqueous  
5 solubility of the chemical or its solubility in the 30 percent  
6 sugar solution or something to that degree.

7 At some point, solubility will define an upper  
8 bound. Perhaps that's something that you guys thought about.  
9 Essentially, I'm asking, is there anything that went into your  
10 thought processes in that regard?

11 **MS. KRISTINA GARBER:** We didn't directly  
12 consider using the solubility. I would think that given that  
13 the nectar would have more than just water, it might complicate  
14 the way that we address solubility, but that's something that we  
15 could consider in the future. But we didn't consider that as an  
16 alternative to the Briggs model.

17 **DR. THOMAS POTTER:** I wasn't referring to the  
18 Briggs model, per se, but in terms of contact exposure from  
19 foliar sprays that are intercepted by pollen and nectar. You  
20 had several studies that you pulled from the literature where  
21 there were some measured data.

22 What was interesting about those studies, and  
23 I'll comment on them in the question period, but they were  
24 looking at -- a couple of the chemicals that were extremely  
25 insoluble in water. So you wouldn't expect to be finding them  
26 in the nectar.



1 I had some concerns about one of the  
2 particular studies. Also about their analytical techniques as  
3 well. But I think it's an important consideration and without  
4 too much effort, I think you could put some thought into that  
5 and bring that into the process. I think it would strengthen it  
6 and certainly add to its credibility.

7 **DR. THOMAS STEEGER:** I would just like to add,  
8 part of the risk assessment process that we'll be talking about  
9 later on this afternoon is the risk characterization component.  
10 Part of that is the risk discussion and just risk description  
11 section of risk characterization and in there the risk assessor  
12 would attempt to capture solubility issues and how it relates to  
13 screening level assumptions so that those types of uncertainties  
14 are indeed characterized in our risk assessments.

15 **DR. THOMAS POTTER:** What you're saying is  
16 you're catching that downstream then?

17 **DR. THOMAS STEEGER:** To the best of our  
18 ability.

19 **DR. THOMAS POTTER:** Okay.

20 **DR. THOMAS STEEGER:** Okay. Thanks.

21 **DR. DANIEL SCHLENK:** Dr. Sandy.

22 **DR. MARTHA SANDY:** I have a question about the  
23 estimate of contact exposures for the foliar spray applications.  
24 So you were basing that on a study with two crops, apples -- I  
25 have to admit I don't know what that other crop was, phacelia or  
26 whatever. So I'm wondering, how representative are those two



1 crops to other crops, like corn and maybe there are others that  
2 bees might come into more contact in the process of gathering  
3 nectar and pollen.

4 **DR. THOMAS STEEGER:** The use of phacelia, it's  
5 a European plant and it has come into considerable use, as of  
6 late, in the semi-field studies, intent on doing screening level  
7 studies is to use a crop that is particularly pollinator  
8 attractive and is a good source of both pollen and nectar.

9 So phacelia fits into that category well. It  
10 has a robust bloom. So presumably, based on it being pollinator  
11 attractive, it is, as I say, presumed to be relatively  
12 protective and a source of information regarding residues that  
13 could accumulate in pollen and nectar, either through a foliar  
14 application or because of systemic transfer to pollen and  
15 nectar.

16 **DR. DANIEL SCHLENK:** Dr. McManaman.

17 **DR. JAMES MCMANAMAN:** I have a question of  
18 clarification, basically because I'm not a bee physiologist, but  
19 your pesticide dose used for the larvae, you claim that the  
20 pollen and nectar have pesticide concentrations, about 100 times  
21 higher in pollen and nectar compared to royal jelly. Is that  
22 correct?

23 **DR. THOMAS STEEGER:** That is correct.

24 **DR. JAMES MCMANAMAN:** I think that your Tier  
25 I, for larvae testing, you're planning on using Day 5 larvae.  
26 What I've read from the literature over the course of the past



1 couple of weeks is that Day 5 larvae still consume royal jelly.  
2 I mean, it certainly would be more conservative, but would that  
3 be inaccurate using those levels to test, would that actually  
4 give result that really wouldn't be applicable to the real  
5 world?

6 **MS. KRISTINA GARBER:** I think that is an  
7 uncertainty associated with the proposed approach that we are  
8 assuming that they would be eating honey and pollen only. We  
9 didn't have data available to quantify how much royal jelly they  
10 may be consuming at that time.

11 So as a conservative approach, we chose to  
12 represent the entire diet as pollen and nectar. So that may  
13 over-predict, but honestly, we don't have any data to tell us  
14 how much.

15 **DR. JAMES MCMANAMAN:** Of the bee experts in  
16 the room, do they consume royal jelly or do they consume pollen  
17 at Day 5 larvae?

18 **UNIDENTIFIED SPEAKER:** Brood food.

19 **DR. JAMES MCMANAMAN:** Pollen? Brood food.  
20 Okay. All right. So it's accurate then. Good.

21 **DR. DANIEL SCHLENK:** Dr. Hunt.

22 **DR. GREG HUNT:** Maybe this will be clarified  
23 later, but with -- it's suggested to use .29 micrograms for seed  
24 treatment products. For clothianidin, that represents about 70  
25 times the LD50 for the smallest estimate of the LD50.

26 Given that this is already being used in corn,



1 I assume that means that it needs to go to Level II trials and  
2 Level III trials. I'm wondering, what are the ramifications?  
3 How long will that take? I'll just throw that out there for  
4 whoever wants to respond.

5 **DR. THOMAS STEEGER:** Without getting into how  
6 this actually plays out in terms of data requirements, as you've  
7 pointed out, for a chemical that has failed a Tier I screen,  
8 first, the decision has to be made by the risk manager whether  
9 to ask for additional information. And the process is  
10 management-based process as opposed to a risk assessment-based  
11 process.

12 So the higher tier studies, it depends on the  
13 nature of the study and it depends on the results, how far you  
14 move up in the refinement process. I don't want to digress too  
15 much here, but depending on the risk assessment question or the  
16 risk management question that's being asked at the higher tier  
17 will dictate the length of the study, particularly if you want  
18 to know what effect overwintering; what effect the chemical may  
19 have overwintering, these can be long studies.

20 The bottom line is that we can't rush the  
21 science to derive an answer that involves regulating pesticides  
22 at a national level. The foundation of our risk assessments has  
23 to be quality science. That being said, it can take a while for  
24 quality science to be generated. I know that's frustrating, but  
25 that's the reality that we have to deal with in terms of being a  
26 regulatory agency that's founded on quality science.



1                   **DR. DANIEL SCHLENK:**       Okay. Any other  
2 questions or clarification on the presentation?

3                   (No response.)

4                   Okay. Let's go ahead with the next  
5 presentation which will be by Reuben Baris.

6                   **MR. REUBEN BARIS:** Good afternoon. My name is  
7 Reuben Baris. I'm an environmental scientist in the  
8 Environmental Fate and Effects Division of the Office of  
9 Pesticide Programs. Thank you very much for the opportunity to  
10 address the SAP.

11                   I will be talking about methods for  
12 characterizing Tier I exposures to bees, as well as study design  
13 and objectives for Tier II exposure studies. These topics are  
14 presented in detail in Section III, and in part, Section IV of  
15 the White Paper.

16                   The studies that I will present in the first  
17 part of this presentation are currently included in the Part 158  
18 data requirements. With minor adjustments and modifications,  
19 these studies have the potential to be used to refine exposure  
20 estimates.

21                   As discussed previously, the risk assessment  
22 process is intended to be iterative. At the Tier I level,  
23 exposure value is used to estimate risk quotients are intended  
24 to be conservative. However, for chemicals identified to be of  
25 concern at the first tier, refined estimates of exposure may be  
26 needed to provide more realistic understanding of pesticide



1 exposures to bees under actual use conditions.

2 Moving from Tier I to Tier II is a function of  
3 risk management needs and decisions. Tier II studies can be  
4 solely measures of exposure as pollen and nectar or can be  
5 coupled with measures of exposure and effects. Higher tiered  
6 studies on the effects of pesticides on honey bees will be  
7 presented by Mr. DeCant, following this presentation.

8 I would like to return your focus to the  
9 proposed decision tree, specifically, seen here in Box 6,  
10 Refining the Tier I Exposure Assessment. I will touch on a few  
11 studies that are currently available, where with modern  
12 modifications to the protocols could be used to refine Tier I  
13 exposure estimates and recalculate risk quotients. The effects  
14 data will not change, but with these studies, we are able to  
15 refine estimates of dietary exposure.

16 For pesticides, they are applied as a foliar  
17 spray and exceed levels of concern for dietary exposure to bees.

18 It may be possible to refine dietary-based estimates of  
19 exposure.

20 This refinement could be accomplished using  
21 pesticide-specific measurements of treated foliage taken within  
22 24 hours of application. Registrant-submitted magnitude of  
23 residue studies used by the Health Effects Division may be used  
24 for this type of refinement.

25 The empirical studies presented on the next  
26 slide may contain information on the transport of a compound in



1 the plant. That is, systemic or non-systemic transport;  
2 therefore, will influence the need for higher tiered studies.  
3 For example, if the chemical in soil is applied and not detected  
4 in plant tissue, fruit, or flower, it is not likely systemic.  
5 Therefore, exposure is unlikely and higher tiered studies would  
6 not be required.

7 The table displayed here outlines empirical  
8 data where identified to be potentially useful for refining Tier  
9 I exposure estimates. These data are routinely submitted as  
10 part of registration or could be submitted if conditionally  
11 required. The Xs represent which study could be used to refine  
12 Tier I estimates of exposure, based on the application type.

13 None of the studies listed are specifically  
14 designed to measure high-end pesticide concentrations in pollen  
15 and nectar at times when bees may be exposed. With modification  
16 to the study design, these studies may be used to generate  
17 chemical-specific measures of pesticides in pollen and nectar  
18 that could be used for chemicals that do not pass the initial  
19 Tier I screen. Some of these studies may be used as refinements  
20 for seed treatments as well.

21 With some limitations recognized, the studies  
22 listed here are currently Part 158 data requirements for  
23 registering pesticides and may be used, with minor  
24 modifications, to study protocols to obtain Day 0 data, or at  
25 the very least, could be used as confirmatory data for  
26 determining the systemic nature of the compound in plants.



1                   Again, I would like to bring your attention  
2 back to the proposed decision tree. For the second portion of  
3 my talk, I will be referring to Box 9(a), Conducting Tier II  
4 Exposure Studies. Box 9(a) is intended for chemical-specific  
5 residue studies as presented earlier by Mr. Sappington.

6                   The purpose of Box 9(a) and the intent of the  
7 Tier II Exposure Assessment is to obtain pesticide-specific,  
8 empirically-based exposure data that represent concentrations  
9 encountered by bees. Like the Tier I exposure method, potential  
10 exposures should be assessed through quantification of doses  
11 through direct spray or diet.

12                   Specifically, studies should be designed to  
13 quantify pesticide residues in pollen and nectar on bees  
14 foraging in the treated field. The dotted line depicted in this  
15 figure shows the process where the Tier II exposure values will  
16 be used to refine Tier I exposure estimates for adults and brood  
17 and then recalculate risk quotients.

18                   Tier II exposure studies with bees are not  
19 typically conducted in the laboratory, but rather focus on  
20 relatively controlled field studies where the movement of bees  
21 to and from the colony is restricted through the use of  
22 enclosures containing the treated crop. These studies may be  
23 designed to measure residues on parent only or parent and  
24 potential degradates of concern.

25                   The next few slides describes the basic  
26 objectives and design elements of the two types of field studies



1 that can be used to quantify pesticide exposure and matrices  
2 that are relevant to bees, such as targeted field studies and  
3 semi-field tunnel studies.

4 The higher tiered studies are designed to be  
5 conducted under vulnerable targeted conditions to provide  
6 quantifiable, refined estimates of pesticide concentrations in  
7 bee food sources.

8 The strength of basing the Tier II exposure  
9 approach on empirical data from field studies is that some of  
10 the uncertainties associated with the Tier I exposure method are  
11 reduced or eliminated. For instance, the assumption  
12 incorporated into the Tier I Exposure Assessment for foliar  
13 applications that pesticide concentrations are in grass are  
14 equivalent to nectar and pollen, is not necessary if pesticide  
15 concentrations are quantified directly in pollen and nectar.

16 Also, the Tier I use of upper bound exposure  
17 values is based on a large set of chemicals that is not  
18 necessary for pesticide-specific residues, can be estimated  
19 under field conditions that are representative of the  
20 pesticide's intended use pattern.

21 As part of the tiered testing process for  
22 evaluating potential exposures and effects on bees, higher  
23 tiered refinements shift the focus from exposure and effects on  
24 individual bees to that of the intact colony. Typical designs  
25 of semi-field studies include a crop that is grown outdoors in  
26 an enclosed system with controlled or confined exposure.



1           The test crop is grown under good agricultural  
2 practices, receiving the maximum-labeled application rate, with  
3 a minimum interval between applications. The test crop provides  
4 the source of pollen and nectar. The design could be structured  
5 to reflect a desired exposure system or foraging environment  
6 such as the mixture of crops and weeds, or flowering margins,  
7 although, such designs can introduce additional variables which  
8 may be difficult to control across replicates and treatments.

9           As is evident, there are a range of different  
10 designs and the actual test structure and study design will  
11 depend on the specific questions asked in the risk assessment in  
12 coordination with risk management needs. Ultimately, the Tier  
13 II exposure studies must be conducted in a manageable way so  
14 that data is collected and used to inform the risk assessment.

15           The field and tunnel residue studies can  
16 provide important information regarding the levels of  
17 contamination potentially encountered by bees in the field;  
18 however, there are a number of uncertainties associated with  
19 these types of studies. One major uncertainty is that the level  
20 of residues inside the hive may be different than those first  
21 encountered by foraging bees in the field.

22           In other words, pesticide concentrations in  
23 freshly collected nectar and pollen could differ from residues  
24 and honey and bee bread. Bee bread and honey, which form the  
25 basis of the hive diet, undergo processing by bees.  
26 Consequently, measures of residues in pollen and nectar



1 collected directly from the flower or from returning bees may be  
2 different from the exposure to individuals within the hive.

3 Tier I exposure estimates are conservative for  
4 unprocessed food sources. The objective of the higher tiered  
5 studies for exposure are dependent upon the Tier I assessment  
6 and where we have the ability to refine those estimates.

7 Refinements are chemical specific  
8 concentrations in pollen and nectar that are used to refine the  
9 quantitative risk quotient. Another notable limitation for the  
10 Tier II Exposure Assessment Method is that it will require a  
11 substantial amount of time and resources to complete.

12 In the future, it may be possible that the  
13 data required from targeted monitoring studies may be used to  
14 establish patterns of exposure based on application methods,  
15 crops and location. These patterns could then be used to  
16 develop a refined method where the data could be extrapolated to  
17 other chemicals.

18 In addition, some field and tunnel residue  
19 studies that are currently being conducted also involve the  
20 collection of data on residues and leaves and flowers. These  
21 data potentially provide a more complete dataset from which to  
22 evaluate the ability of flowers or leaf residues to provide a  
23 conservative measure of residues in pollen and nectar.

24 The first type of Tier II study presented is a  
25 targeted field study or field residue trial, where pesticide  
26 residues are quantified in pollen and nectar, collected directly



1 from the treated crop. This study could be specifically  
2 designed with the sole intent of quantifying pesticide residues  
3 and matrices relevant to honey bees, or could be incorporated  
4 into the study design of an existing protocol required for  
5 pesticide registration, such as a cropped terrestrial field  
6 dissipation study.

7 In the design of these studies, it is  
8 important to consider the type of application, that is, foliar,  
9 soil, or seed treatment and relate the design to the desired  
10 outcome.

11 The second study is referred to a tunnel study  
12 because the treated crop is kept under an enclosure, along with  
13 a nucleus hive of bees, which are small colonies with roughly  
14 5,000 bees. Study design guidance is currently provided by  
15 EPPO, OACD, and also more recently by EFSA.

16 The selection and use of this study, compared  
17 to the targeted field study, must be considered, along with the  
18 desired outcomes of the Tier II study. For example, if the  
19 desired outcome is to collect pollen, nectar, and bee residue  
20 data, that is, pollen sacs, pollen traps, or regurgitated  
21 nectar, then the semi-field study would meet those objectives.

22 If the goal is to obtain targeted field data,  
23 that is, measured residues in pollen and nectar directly from  
24 plants, the targeted field study would meet those objectives.  
25 The difference in the goals of the Tier II studies is the  
26 measured residues of relevant bee matrices, where the tunnel



1 study design is set up to measure bees and hive products.  
2 Additional detail for the Tier II study design will be  
3 presented, following this presentation.

4 There are several sources of variation related  
5 to the anticipated use of the pesticide that should be  
6 considered when designing the studies for Tier II Exposure  
7 Assessment, including application methods, props, and location.  
8 Application methods employed for a chemical can vary. Because  
9 different application methods may result in different levels and  
10 types of exposure application practices that are expected to  
11 generate high-end exposure concentrations for pesticides should  
12 be selected.

13 In determining which crops to include in field  
14 studies, a focus should be placed on registered uses for crops  
15 that are highly attractive to bees and that have the highest  
16 application rates. For systemic pesticides in particular,  
17 different plant species may have differences in uptake,  
18 distribution, and metabolism of the test substance.

19 In addition, different plants have different  
20 matrices that represent food sources for bees. For example,  
21 corn only produces pollen that bees will collect as a food  
22 source, whereas, canola produces both nectar and pollen. Some  
23 plants, such as cotton, also produce extra floral nectaries  
24 which may be present when flowers are not.

25 The sampling scheme is an important  
26 consideration in the design of the Tier II studies. Sampling



1 should target the maximum concentrations found in pollen and  
2 nectar, therefore, these studies should sample pollen and nectar  
3 at different time points during bloom to evaluate the presence  
4 and decline of the compound over time.

5 Sample timing should be selected based on the  
6 consideration of the dissipation of the chemical through  
7 degradation and transport, away from the plant, as well as  
8 systemic transport through the plant to pollen and nectar. For  
9 foliar applications, studies should be designed such that the  
10 pesticide concentrations are quantified on the day of  
11 application and during subsequent days that are selected to  
12 represent the concentrations on bees and relevant plant samples.

13 For soil and seed treatment applications,  
14 sampling should account for the delayed transport from roots to  
15 pollen and nectar. Different field locations have different  
16 climates and soils, and therefore, growing conditions at a  
17 spacial scale.

18 The locations of the studies should be  
19 selected so that they represent anticipated use sites of the  
20 chemical. If possible, honey bee specific studies should be  
21 conducted in multiple regions that are identified to be of  
22 concern.

23 As a result of these considerations, it may be  
24 necessary to conduct targeted studies, using multiple locations,  
25 methods, crops, and locations in order to quantify potential  
26 pesticide exposures to bees under conditions that are



1 represented of the anticipated use of the chemical.

2 Because field residue data may exhibit extreme  
3 spacial and temporal variability, it is necessary to collect a  
4 sufficient amount of data to allow the risk assessor to have an  
5 understanding of the central tendency and upper bound exposure  
6 that represents the potential environmental exposures. Thank  
7 you very much for your time. I would like to open the floor to  
8 any clarifying questions the panel may have.

9 **DR. DANIEL SCHLENK:** Any questions  
10 regarding exposure. Again, we're going to be talking about  
11 toxicity in the next presentation. Dr. James?

12 **DR. ROSALIND JAMES:** Did you take into account  
13 the formulation at all? Is it at this point? Do you ever take  
14 into account formulation?

15 **DR. THOMAS STEEGER:** At a screening level, we  
16 have available to us formulation toxicity data that are  
17 submitted for all different types of formulations. And from  
18 that, we get a sense for the extent to which different  
19 formulations may be toxic - more toxic than the technical-grade  
20 active ingredient.

21 To be candid, those data are on mammals. If  
22 we have information for existing chemicals that particular  
23 formulations may be more toxic than a technical-grade active  
24 ingredient based on open literature data or an incident data, we  
25 have the purview to require studies looking specifically at  
26 those particular formulations.



1                   **DR. ROSALIND JAMES:**   There's a toxicity  
2 question, but then also it might affect exposure, like one of  
3 the famous cases of microencapsulation where the bees were going  
4 after the microcapsules if they were pollen, so they were  
5 actually having a higher exposure. So to me, it seems like Tier  
6 II might be a place where that came into play, but I wasn't sure  
7 if you had considered that.

8                   **DR. THOMAS STEEGER:**       The microencapsulated  
9 products, particularly like Penncap-M that had particular affect  
10 on bees, a lot of that information became available to us  
11 through incident data. As we'll talk about it in my  
12 presentation, incident data serves as a means for the Agency to  
13 sort of ground truth its risk assessments with an understanding  
14 of how that chemical is performing under actual use conditions.

15  
16                   So it's a critical need of ours to have access  
17 to that information as well as open literature information that  
18 would also inform us. But you're correct; there are  
19 formulations that you would be able to better glean through a  
20 higher tier testing.

21                   **DR. DANIEL SCHLENK:**       Dr. Potter.

22                   **DR. THOMAS POTTER:**   I have a quick question.  
23 Have you had any studies of this type, the Tier II type or the  
24 Tier III type, submitted for your review?

25                   **DR. THOMAS STEEGER:**       Yes, we have.

26                   **DR. THOMAS POTTER:**   How did it go?



1                   **DR. THOMAS STEEGER:**           I think that the  
2 higher tier studies, to date, have been difficult to interpret  
3 because they tend to ask or look at very broad questions. One  
4 of the things that we're proposing in the White Paper is that as  
5 you transition through the various tiers of refinement, the  
6 questions that are being asked are intended to address specific  
7 uncertainties and that we get away from shotgun approaches to  
8 doing field studies to those that are answering very, very  
9 refined question. I'm hoping that through that process it'll be  
10 much easier to interpret the results of those studies.

11                   **DR. DANIEL SCHLENK:**           Something to add?

12                   **MR. KEITH SAPPINGTON:** I would like to just  
13 add that based on my experience in looking at a number of field  
14 studies, first of all, in those studies, the typical end-use  
15 product was used so we did have a sense of the different  
16 formulations.

17                   But secondly, and this is, I believe, part of  
18 the charge questions, is understanding biological significance,  
19 visa-vis, statistical significance because we have difficulties  
20 with statistical power. It is important, even more, I think, to  
21 have a good sense of what is biologically significant from  
22 different endpoints.

23                   **DR. DANIEL SCHLENK:**           Any other -- Dr.  
24 Tarpy.

25                   **DR. DAVID TARPY:** Based on Figures 2 and 3 in  
26 the White Paper, if a particular compound is shown in Tier I not



1 to have any concern, then it does not go on to Tier II or III by  
2 default, correct?

3 **DR. THOMAS STEEGER:** As Steve Bradbury  
4 pointed out earlier, the risk quotients that are calculated at a  
5 screening level are not viewed in and of themselves. We take  
6 into consideration other lines of evidence that might be  
7 available, and those would include open literature studies that  
8 might've been conducted; field studies that the registrant may  
9 have conducted as part of their testing process, and again,  
10 incident data.

11 It's those other lines of evidence that if we  
12 feel are in accordance with our understanding of the lack of  
13 risk at a screening level, then that would be correct. But if  
14 there is information to suggest otherwise, then it would be up  
15 to the risk manager to determine whether higher tier levels of  
16 refinement would be needed.

17 **DR. DAVID TARPY:** Have there been examples of  
18 compounds at Tier I have shown no concern, but tested that Tier  
19 II and actually do have effects at that level?

20 **DR. THOMAS STEEGER:** I don't want to cite a  
21 particular chemical, but I think that some of the current  
22 limitations in the testing process where we don't do -- we  
23 haven't done larval toxicity testing as a matter of routine, and  
24 compounds that can be completely innocuous to adult bees, but  
25 quite detrimental, such as insect growth regulators to the  
26 larval stage, might make it through that screen but now we are



1 proposing a means to address both life stages.

2 **DR. DAVID TARPY:** Thank you.

3 **DR. DANIEL SCHLENK:** Okay. Any other  
4 questions before we break? Let's try to be back about five 'til  
5 3:00. 2:55.

6 (Brief recess.)

7 **DR. DANIEL SCHLENK:** Okay. Our first  
8 presentation is by Joseph DeCant.

9 **MR. JOSEPH DECANT:** Good afternoon. My name  
10 is Joseph DeCant, and I'm an ecologist in the Environmental Fate  
11 and Effects Division. I'll be talking about the various methods  
12 that are available for evaluating the toxicity of pesticides to  
13 bees, as outlined in the White Paper.

14 Throughout this talk, I intend to present our  
15 studies. The first step, though, will be to provide an overview  
16 of how these toxicity studies relate to each other and the risk  
17 assessment process, and the overall tiered approach to  
18 toxicity evaluation.

19 This talk will also encompass the linkages  
20 between the different tiers and introduce the triggers that  
21 provide the foundation for moving from one tier to the next  
22 within the toxicity-testing framework. Finally, I would like to  
23 call your attention to Charge Questions 8 through 12, which this  
24 presentation will touch on and to which it will provide context.

25  
26 In order to understand the importance of the



1 toxicity studies themselves and their relation to the risk  
2 assessment process, I want to recall Table 1 from the White  
3 Paper, regarding the various protection goals. Note, that for  
4 each protection goal, there are associated assessment endpoints  
5 that will be evaluated in the risk assessment.

6 In addition, there are also associated  
7 measurement endpoints that will be evaluated through certain  
8 toxicity studies. Therefore, keep in mind that each of the  
9 toxicity studies available for the risk assessment should  
10 provide one or more measurement endpoints with a clear linkage  
11 to an assessment endpoint of concern.

12 As we have already heard in the previous  
13 presentations, we have explored methods relating to evaluating  
14 exposure, both at the screening level and more refined methods  
15 that provide information and actual residues in the crops of  
16 interest.

17 The goal of these refinements has been to  
18 capture more realistic measures of exposure, albeit at greater  
19 expense in terms of the cost of conducting and reviewing such  
20 studies. The approach is similar with toxicity studies and  
21 their use within the risk assessment process.

22 Consequently, the tiered approach to toxicity  
23 testing is meant to provide a means of evaluating the  
24 measurement endpoints described in the previous slide through an  
25 iterative evaluation process that reflects a progressively more  
26 realistic environment and refines the earlier tier evaluations.



1           For example, at the screening level, toxicity  
2 studies are based on the individual in a laboratory; however, as  
3 we move through Tiers II and III, we move to studies that  
4 evaluate the entire colony and eventually in the field under  
5 actual application scenarios for specific labeled uses.

6           For the first year, the U.S. EPA 40 CFR Part  
7 158 data requirements currently require an acute contact  
8 toxicity test with young adults identified as guideline  
9 850.3020. In addition to this test, Canada also requires an  
10 acute oral toxicity test with young adult bees.

11           A recent EFED Guidance document, the Interim  
12 Guidance on Honey Bee Data Requirements, provided an overview  
13 of various studies for use of the screening level and included  
14 both the acute contact and oral studies with young adult bees  
15 and in an in vitro study for the larval stage of honey bees.

16           Similarly, the proposed approach presented in  
17 the White Paper expands upon the current U.S. EPA requirement of  
18 the acute contact study to include these two additional oral  
19 exposure studies. This expansion is consistent with the  
20 USDA-led workshop, as well as two recent publications; one from  
21 a 2011 SETAC Pellston Workshop, as well as one from the European  
22 Food Safety Authority in 2012.

23           Through the expanded Tier I battery of  
24 toxicity tests, we can evaluate the toxicity of a pesticide for  
25 both contact and dietary routes of exposure. I also want to  
26 mention at this time that these Tier I toxicity tests will be



1 used to evaluate the stressors of concern to honey bees.  
2 Therefore, in some cases, degradates may be identified to be of  
3 concern as well. In these cases, toxicity tests will evaluate  
4 the parent compound as well as any potentially toxic degradates.

5  
6 A U.S. EPA and OECD Guideline evaluate acute  
7 contact toxicity to young adult honey bees. The study produces  
8 a 48-hour LD50 or a lethal dose at which 50 percent of the test  
9 bees dies. The study can be expanded to 96 hours if it is  
10 appropriate, as in the case of chemicals that showed delayed  
11 toxicity.

12 The most sensitive LD50 values are intended to  
13 be used in combination with the appropriate estimated exposure  
14 value to derive risk quotients for the Tier I risk assessment.  
15 In cases where a pesticide is not applied by foliar spray, the  
16 acute contact toxicity data may not be necessary for assessing  
17 the risks of a chemical because for soil applications, seed  
18 treatments and tree trunk applications, dietary exposure is  
19 expected to be the predominant route.

20 Depending on the outcome of these toxicity  
21 tests, pesticides are classified as practically non-toxic,  
22 moderately toxic, or highly toxic to bees on an acute exposure  
23 basis. If the acute contact LD50 is less than 11 micrograms of  
24 active ingredient per bee, additional testing may be required in  
25 the form of a foliar residue study to determine the duration  
26 over which field weathered foliar residues remain toxic to honey



1 bees.

2 This guideline study, 850.3030, provides an  
3 RT25 or the time at which a chemical produces 25 percent or less  
4 mortality. The RT25 will not be used to quantitatively evaluate  
5 risk, but rather the study will provide another line of evidence  
6 related to the contact toxicity of residues on foliages over  
7 time.

8 Although not currently required by EPA, Canada  
9 and the European Union routinely require acute oral toxicity  
10 data for adult worker bees. The acute oral toxicity study is a  
11 laboratory test method designed to assess a mortality of young  
12 adult worker bees, following a single oral dose of a pesticide.

13  
14 A standard OECD Guideline exists for this  
15 study. Similar to the acute contact toxicity test required by  
16 EPA, the study provides a 48-hour LD50 value and can be extended  
17 to include a 96-hour LD50 if mortality is demonstrated to  
18 increase between 24 and 48 hours.

19 The proposed Tier I Risk Assessment Methods  
20 involves using the 48-hour or 96-hour LD50 generated from this  
21 study to derive risk quotients representing risk to adult bees  
22 exposed to pesticides through the diet, following foliar spray  
23 application, soil application, seed treatments, and tree trunk  
24 treatments of pesticides.

25 The previous two slides show the studies that  
26 will provide measures of toxicity to adult honey bees. Unlike



1 with the acute contact in oral toxicity studies for adult worker  
2 bees, established regulatory test guidelines for assessing  
3 larval toxicity do not exist. Given that larvae may be exposed  
4 to pesticides through the diet, the lack of understanding of the  
5 toxicity of a chemical on larvae is considered to be a  
6 significant data gap.

7 Therefore, efforts are underway to identify  
8 critical design elements of an acute oral larval toxicity study  
9 that may be used to understand the toxicity of a chemical to  
10 this lifestage of worker bees. One option is a field-feeding  
11 design that looks at impacts to the brood at the whole hive  
12 level. This study is considered a semi-field design and will be  
13 discussed later in the presentation.

14 Another option is an in vitro assay based on  
15 the method, developed by Aupinel et al., with larval honey bees,  
16 which will provide a screening evaluation of toxicity to  
17 immature stages.

18 In this method, first instar bee larvae are  
19 transferred into 48-well plates and fed a synthetic royal jelly  
20 sucrose solution mixture containing known quantities of the test  
21 chemical over the 48-hour period.

22 This study can provide an LD50 but it can also  
23 potentially provide a no effect concentration, hereafter  
24 referred to as NOAEC, if there is sufficient replication.

25 The methodology has been ring tested and a  
26 review indicated that additional adjustment in the method may be



1 needed to adjust potential sources of variability, including  
2 colony origin of the brood, season and larval heterogeneity  
3 grafting. It is also important to note that the study design  
4 has a number of uncertainties associated with it, including  
5 laboratory conditions in which the larvae must survive outside  
6 of the hive environment.

7 In addition, the larvae are directly fed an  
8 artificial mixture of royal jelly and sucrose solution, which is  
9 different from what the larvae would be fed, after processing,  
10 by adult nurse bees in the hive.

11 The studies that I have discussed so far are  
12 meant to evaluate acute toxicity. Chronic laboratory-based  
13 toxicity testing has not been routinely required on individual  
14 bees in the U.S.; rather information on the potential chronic  
15 toxicity of pesticides to bees has historically been discerned  
16 from whole colony studies conducted under the semi-field or full  
17 field conditions.

18 The proposed Tier I risk assessment method for  
19 bees includes quantification of the toxicity of larvae and adult  
20 worker bees following chronic exposures to pesticides; however,  
21 no formal guidelines have been developed, to date, for  
22 conducting chronic toxicity tests with either adult or larval  
23 bees.

24 Efforts are underway to identify design  
25 elements of chronic larval and adult toxicity studies that may  
26 be used to understand the toxicity of a chemical to worker bees



1 and test protocols already exist for these types of studies. A  
2 10-day oral chronic toxicity test with young adults would be  
3 used to quantitatively evaluate risk to adults.

4 The USDA technical working group report noted  
5 that this type of chronic toxicity test with individual adult  
6 bees has already been conducted to support registrations in the  
7 EU. These tests consist of collecting newly emerged juvenile  
8 bees from the comb and placing 50 of these bees per cage.

9 A sufficient number of replicates are run,  
10 along with suitable negative and reference controls to conduct  
11 hypothesis testing to establish a NOAEC and LOAEC. The bees are  
12 fed a sucrose solution; however, no protein source, such as  
13 pollen, is provided.

14 Exposure is through spikes made to the sucrose  
15 solution and cages of bees are provided 10 milliliters of test  
16 solution every other day. Bees are observed for a total of 10  
17 days, and mortality is a primary measurement endpoint. The in  
18 vitro chronic larval toxicity study is similar in design to the  
19 previously mentioned acute larval toxicity study, though with a  
20 longer duration of feeding and monitoring.

21 It should also deliver in NOAEC as well when  
22 monitoring of toxic effects is extended to seven days; however,  
23 at this point, the chronic adult and larval study methods  
24 require further evaluation prior to use in a regulatory setting.

25 Another approach is to derive a no effect  
26 concentration from an acute toxicity study; however, because it



1 is a regression-based study targeted to ascertain the LD50, the  
2 dosing regime is not intended to assess the no effect level.  
3 Consequently, there is uncertainty as to the actual  
4 concentration where no effects occur.

5 Hypothesis-based testing in chronic studies is  
6 more appropriate as the dose election targets the low  
7 concentrations around the no effect level. Additional data are  
8 often available that may be useful in characterizing the effects  
9 of a pesticide in bees.

10 The first major bullet notes scientific  
11 literature which often includes toxicity data involving bees.  
12 Acute toxicity data for honey bees may be available in  
13 scientifically valid studies from the open literature, for  
14 example, peer-reviewed journal articles. But these studies from  
15 the open literature must be sufficiently robust, based on EFED  
16 open literature review guides.

17 In some cases, these values may be used to  
18 derive risk quotients when these endpoints are lower than those  
19 found in registrant-submitted studies.

20 The second major bullet point relates to data  
21 on non-target arthropods, other than bees. These data are not  
22 used to derive risk quotients, but are considered in the effects  
23 characterization. For example, data are often available from  
24 registrant-submitted acute toxicity studies involving other  
25 non-target arthropods, such as the green lacewing and parasitoid  
26 wasps.



1           The final major bullet point addresses reports  
2 of honey bee mortality events -- that is, bee kill incident  
3 reports -- following pesticide exposures. These incident  
4 reports may be useful in characterizing the available risk  
5 estimates from laboratory toxicity data.

6           Although the proposed measurement endpoints  
7 for acute and chronic exposures are based on mortality, there  
8 are an increasing number of sublethal endpoints reported in Apis  
9 and non-Apis toxicity studies, which may be conceptually  
10 relevant to the proposed assessment endpoints.

11           Sublethal effects therefore may also provide  
12 additional lines of evidence in the risk assessment. These  
13 effects are those that do not directly cause the death of an  
14 individual organism and are listed in the slide.

15           The acute and chronic studies may provide  
16 information on sublethal effects, but they were designed to  
17 evaluate mortality and not address concentrations that would  
18 elicit sublethal effects. This is the reason that the EFSA  
19 Scientific Opinion of pollinator risk assessment concluded that  
20 the current battery of toxicity studies are not adequate at  
21 addressing sublethal effects. It recommended that sublethal  
22 effects are important indicators of bee health and vitality and  
23 must be accounted for in the risk assessment process.

24           These endpoints also represent a challenge in  
25 that in most cases, they do not provide a direct connection to  
26 our assessment endpoints. Before using these endpoints to



1 derive risk quotients, the relevancy of these sublethal  
2 measurement endpoints for evaluating assessment endpoints needs  
3 to be established through quantitative links to colony strength  
4 and survival.

5 As the last bullet notes, although such  
6 endpoints may not be used quantitatively to estimate potential  
7 effects on assessment endpoints, studies on sublethal effects  
8 could potentially be used qualitatively to describe effects  
9 which may not have clearly established relationships to colony  
10 level effects or for explaining mechanisms by which colony level  
11 effects might occur.

12 Consequently, these sublethal effects will be  
13 used to qualitatively describe risk in the assessment until  
14 further research has been conducted that establishes a link  
15 between these effects and assessment endpoints.

16 This slide provides a representation as to how  
17 the data will be used in the risk assessment. The endpoints for  
18 quantitative use, which are depicted in the green box, will be  
19 combined with either model estimates of exposure or measured  
20 residue levels in order to develop risk quotients.

21 The information contained in the sources in  
22 the red box will be used qualitatively to further describe the  
23 risk that the risk assessor is quantified. This risk analysis  
24 will then be related to the assessment endpoints of concern,  
25 highlighted in the blue box, and will be used to inform the risk  
26 manager, regarding the screening level of results of the use of



1 a particular pesticide.

2 At the Tier I screening level, effects data  
3 may primarily be based on laboratory studies, while exposure  
4 estimates may be based on models and/or conservative default  
5 values, as well as more refined measures of exposure from field  
6 monitoring studies.

7 These studies are evaluated using relatively  
8 conservative assumptions to provide high-end estimates of  
9 potential risk such that if a chemical passes a screen, the  
10 presumption of low risk to non-target organisms is considered,  
11 in most cases, protected.

12 As part of the tier process, there's a  
13 progression from the left box to the right box in that higher  
14 tiered studies are intended to reflect increasing levels of  
15 realism, albeit from a less controlled testing environment, in  
16 terms of how organisms may be exposed and the nature of the  
17 effects which results from such exposure.

18 Also, as part of the tier testing process,  
19 higher tiered refinements shift the focus from exposure and  
20 effects on individual bees to that of the intact colony. The  
21 social nature of honey bees requires extensive interaction and  
22 communication among various castes of bees for successful  
23 development and propagation of colonies.

24 As a result, numerous linkages exist between  
25 pesticide exposure and effects on individual bees and their  
26 collective impact on honey bee colony health, which are not



1 readily assessed in the laboratory.

2           Additionally, the Tier II semi-field studies  
3 provide a means of assessing both exposure and effects that are  
4 less conservative and more realistic than the methods used in  
5 laboratory testing. It is important to note that Tier II  
6 studies address the limitation of the Tier I screening level  
7 related to sublethal effects, chronic exposure and potential  
8 degradate toxicity by integrating these factors into the colony  
9 level measurement endpoints.

10           Thus, Tier II studies may be conducted if  
11 concerns remain from Tier I levels of the risk assessment and  
12 further information is needed for a more refined estimate of  
13 potential risk to honey bees. Ultimately, the transition from  
14 Tier I to Tier II is based on more than just the relationship  
15 between exposure and toxicity.

16           In the case of a chemical that either passes  
17 or fails a screen, the risk manager has a number of different  
18 types of information available, including the previously  
19 described toxicity and exposure data, other lines of evidence,  
20 such as incidents, uncertainties related to the characteristics  
21 of the chemical itself and whether the Tier I analysis has  
22 substantial gaps and knowledge, potential mitigation options,  
23 and finally, the option to off-label specific uses.

24           Consequently, Tier II studies are based on the  
25 needs of the risk manager and are based on information of the  
26 impact of the pesticide, as identified, in lower tier studies.



1                   Unlike the screening level studies, Tier II  
2 studies with bees are not typically conducted in the laboratory,  
3 but rather focused on relatively controlled field conditions,  
4 referred to as semi-field. In these studies, the movement of  
5 the bees to and from the colony is restricted through the use of  
6 enclosures containing crops treated with the pesticide, or  
7 feeding bees a pesticide-spiked diet.

8                   Therefore, a Tier II toxicity study is meant  
9 to refine the Tier I assessment by providing a means of  
10 evaluating the impacts to the assessment endpoints at the whole  
11 hive level. Compared to laboratory studies where exposure is to  
12 an individual organism and typically through a single route,  
13 such as contact or oral exposure, semi-field studies can provide  
14 clear lines of evidence for linking multiple routes of exposure  
15 to adverse ecological effects at the colony level.

16                  Semi-field studies are also useful for  
17 determining the extent to which effects on individual bees  
18 identified in Tier I laboratory studies are expressed at the  
19 colony level. The decision to move to higher tier testing is  
20 also supported by current U.S. EPA 40 CFR Part 158 data  
21 requirements for the testing of pesticides.

22                  The conditions for the transition to higher  
23 tier studies are based on the residual toxicity data that  
24 indicate there's the potential for effects other than mortality  
25 on the colony, and data that indicate potential chronic,  
26 reproductive, or behavioral effects.



1                   Although the current EPA data requirements for  
2 pesticides identified in the 40 CFR Part 158 do not specifically  
3 include semi-field studies, the Guideline 850.3040 on field  
4 testing for pollinators is general and would accommodate such  
5 studies.

6                   The design of the Tier II semi-field study is  
7 based upon the specific issues raised at the initial tiers of  
8 the risk assessment. The specific design is flexible and,  
9 consequently, can be tailored to suit the needs of the risk  
10 assessment.

11                  In general, there are two basic designs of the  
12 semi-field studies. One employs an enclosure, often called a  
13 tunnel, as seen in the top picture of the slide, and can find  
14 small nucleus hives to ensure that the bees forage on a treated  
15 crop. Smaller hives are used in this type of study in order to  
16 prevent the colony from overwhelming the forage base within the  
17 enclosures.

18                  The other design allows bees to freely move,  
19 but provides artificial food sources, as seen by the lower  
20 picture with pollen cake, spiked with a pesticide of interest.  
21 Although the feeding study designs do not involve enclosures and  
22 bees are allowed to forage freely, the studies are considered  
23 semi-field since test organisms are provided pesticide-spiked  
24 food, which does not occur in full field studies. Both designs  
25 offer relatively controlled conditions in comparison to Tier III  
26 full field studies, yet enable whole hive evaluations to be on



1 Tier I screening laboratory studies.

2 As is evident, there are a range of different  
3 designs and the actual test structure will depend on the  
4 specific questions asked in the risk assessment. There are a  
5 number of already established guidelines for the semi-field  
6 tunnel study.

7 The typical designs of semi-field studies  
8 include a highly pollinator attractive crop or surrogate species  
9 such as phacelia that is grown outdoors in an enclosed system  
10 with confined exposure.

11 The test crop is grown under good  
12 agricultural practices using the maximum application rate with  
13 the minimum application interval. The test crop provides the  
14 source of pollen and nectar. The test design employs a tunnel  
15 in which small nucleus hives containing about 5,000 worker bees,  
16 brood in all stages, and stores of pollen and nectar are placed.

17 A test crop is used at or near full bloom that  
18 is representative of high-end exposure condition for the given  
19 type of application method and rate. For foliar sprays, the  
20 hives are introduced in the tunnel two to three days before  
21 pesticide application and hive condition is assessed while the  
22 bees acclimate to the enclosure.

23 Pesticides can then be applied at peak bee  
24 foraging activity to maximize exposure to foraging bees. Hives  
25 are kept in the tunnel for a period of seven to ten days, after  
26 the first pesticide application and allowed to forage on the



1 treated crop. The hives are then removed from the tunnel and  
2 moved to an area of minimal pesticide use to forage on untreated  
3 areas with continued monitoring, which is usually up to a total  
4 study period of 28 days but sometimes longer.

5 A reference chemical is also frequently tested  
6 to demonstrate the application method used that actually results  
7 in exposure to bees, as well as the ability of the study to  
8 detect and quantify effects. Reference chemical selection is  
9 dependent on the expected mode of action of the pesticide under  
10 consideration, which would be a chemical with known toxicity to  
11 the hive.

12 For pesticides that are expected to directly  
13 impact brood development, an insect growth regulator, such as  
14 fenoxycarb, is typically used. For pesticides that are expected  
15 to elicit effects directly on adults and indirectly on brood,  
16 through rapid knockdown, a reference chemical that acts  
17 similarly, such as dimethoate, is often selected.

18 In addition to the monitoring of toxic  
19 effects, residues in pollen and nectar on the flowering crop can  
20 also be provided in order to assess the levels of exposures to  
21 the honey bee colonies.

22 As the last bullet highlights, it is important  
23 to note that this design can be used to evaluate each of the  
24 application methods, depending on when the tunnels are in place.

25 Moving to the feeding designs, one design  
26 typically addresses brood toxicity. The study is based on a



1 method by Oomen, et al. in 1992, which uses hives that can  
2 freely forage in order to test for the possibility of adverse  
3 effects on the brood within a honey bee hive. The basic design  
4 uses one liter of a sucrose solution that is placed in a feeder  
5 near a full-sized honey bee hive. The sucrose solution is  
6 either a control without contamination or a solution spiked with  
7 a known concentration of a pesticide.

8 The solution is provided as a food source and  
9 once the hive has collected all of the solution, weekly  
10 measurements are taken on brood development within the hives.  
11 In addition to the data on brood development, data on adults and  
12 brood mortality estimates can be obtained through counts of bees  
13 collected in dead bee traps, placed at the entrance of the  
14 colony.

15 The test chemical concentrations are not  
16 necessarily meant to be reflective of an environmentally  
17 relevant concentrations, but rather may be exaggerated in order  
18 to screen chemicals for possible impacts to the brood when in a  
19 whole hive situation.

20 The EPA has also explored the modification of  
21 the feeding design to expand it to adults as well as brood  
22 toxicity, to incorporate multiple dietary pathways of exposure  
23 and to increase the duration of the study if necessary. It  
24 should be noted that these refinements have not been well  
25 vetted, but a number of field feeding designs are increasingly  
26 found in the open literature, reflecting the potential for this



1 type of experiment to address uncertainties related to dietary  
2 exposure.

3 For this type of study, spiked food, as in  
4 pollen patties, sucrose solution or both, can be fed ad libitum  
5 to full size honey bee hives of at least 10,000 bees over a  
6 specified duration of time and during a specified time of the  
7 year. Different concentrations of the chemical and food are  
8 used with the intent of identifying a NOAEC and LOAEC.

9 The treatment levels are either selected from  
10 concentrations of concern that have been identified in the body  
11 of literature or by environmentally relevant concentrations  
12 found in targeted monitoring studies for residues in pollen and  
13 nectar. Therefore, the design should be based on  
14 environmentally relevant concentrations in an attempt to  
15 ascertain where the no effect level is found, relative to these  
16 concentrations.

17 The design does not employ tunnels, so the  
18 bees are allowed to forage freely. As the second bullet notes,  
19 while the potential exists for bees to forage in areas where  
20 other pesticides may be in use, test site selection is intended  
21 to minimize the potential for such exposure. Consequently, the  
22 test hives are placed in an area of low pesticide use in order  
23 to minimize any exposure to other pesticides.

24 Related to the third bullet, during a field  
25 feeding study, exposure can be controlled to a greater extent  
26 than a full field study through the use of pollen traps, to



1 limit incoming pollen and encourage feeding on the treated food  
2 source. As previously mentioned, the enclosures used in the  
3 typical semi-field design introduced stress to the colony and  
4 effective brood performance on the colonies in as little as  
5 seven to ten days.

6 Moving to the fourth bullet, the open field  
7 feeding design allows a colony to freely forage, thereby  
8 eliminating the stress of the enclosure. Furthermore, as the  
9 fifth bullet states, the study hives can be large enough to  
10 survive through overwintering, unlike the smaller nucleus hives  
11 typically used in the tunnel studies.

12 The use of larger colonies enables a study to  
13 proceed over a longer duration than other semi-field studies in  
14 an enclosure. With this flexibility, the full field feeding  
15 design can be used to evaluate a colony's response to either  
16 short-term or a long-term exposure condition to evaluate  
17 overwintering performance of the hives.

18 Consequently, this study can address the  
19 uncertainties identified at the lower tier levels or even at the  
20 initial stages of the Tier II toxicity assessment with other  
21 semi-field studies, specifically when oral exposures is the  
22 pathway of concern.

23 Finally, as the last bullet states, the  
24 measurement endpoints can be taken at specified time periods to  
25 evaluate the hive at the most relevant time points, relative to  
26 the nature and concern of the chemical.



1           The results of this study are intended to  
2 reflect to the extent to which specific effects may occur in  
3 response to various pesticide levels in pollen and nectar when  
4 compared to measured residues from targeted monitoring studies.

5           Assuming that targeted and monitoring studies  
6 are available to design the field feeding study or are available  
7 following the completion of the feeding study, these data would  
8 provide a measure of the level of residues in pollen and nectar  
9 from various crops. This feeding study could then provide a  
10 context for these residue data at the whole hive level.

11           Given the assumption that exposure to  
12 concentrations in the pollen and nectar are equivalent to  
13 exposure from concentrations in the sucrose solution and/or  
14 pollen patties, the study would reveal to what extent the risk  
15 assessor could anticipate effects at the residue levels in the  
16 different crops by comparing the concentrations that produce an  
17 effect and measured concentrations in pollen and nectar.

18           The comparisons between the no effect level  
19 and the residue levels are not quantitative comparisons to be  
20 used as triggers, but rather a means of understanding the  
21 potential impacts on the colony, given realistic residue levels  
22 in pollen and/or nectar from various crops.

23           The study results could also further describe  
24 the nature of these impacts to the risk manager to consider with  
25 other lines of evidence. The measurement endpoints are  
26 determined by the protection goals and remaining uncertainties



1 from the lower tiers in coordination with the risk manager.

2 Measurement endpoints include adult bee  
3 mortality, brood condition, hive strength, queen health, and  
4 hive resources which are measured at various points over the  
5 duration of the study.

6 The endpoints should also be specific to the  
7 uncertainties identified earlier in the tiered process as they  
8 relate to the assessment endpoints, so as to minimize the number  
9 of endpoints to be collected because continued opening of the  
10 hives can add stress to the colony.

11 Another major strength of these semi-field  
12 studies is that they integrate effects from toxic degradates,  
13 chronic exposure and sublethal effects. Two prominent  
14 uncertainties at the Tier I screening level are that the  
15 laboratory studies either are inadequate to screen for various  
16 types of sublethal effects and there are no currently  
17 established protocols to evaluate chronic toxicity.

18 The endpoints measured in the semi-field  
19 studies integrate each of the previously mentioned factors and  
20 that have toxic degradates, chronic exposure of sublethal  
21 effects impact these measurement endpoints which are also tied  
22 to assessment endpoints of concern and the study reflects the  
23 sum of both these lethal and sublethal effects.

24 In addition to the strength of the semi-field  
25 studies, it is also important to note that there are limitations  
26 to these study designs as well. Since the studies are conducted



1 outside of a laboratory environment, feeding and tunnel designs  
2 are subject to the changes of weather, as noted in the OECD  
3 Guidance for Semi-Field Studies, the test cannot be performed  
4 under adverse weather conditions. In addition, there is a high  
5 degree of variability in the measurement endpoints, not only  
6 between hives, but within the same hive over time as well.

7 Related specifically to the tunnel design  
8 because bees are not allowed to forage freely over large areas,  
9 the quantity and quality of food they are able to gather may be  
10 compromised, compared to open field conditions. Experience with  
11 these studies indicates that confinement stress on foraging  
12 bees, among other factors, often leads to adverse effects on  
13 egg, larval, and pupil abundance, with increasing duration of  
14 confinement in the tunnels.

15 As a result, hives can only be kept in tunnels  
16 for a relatively short period of time that is seven to ten days,  
17 to ensure adequate condition of controls for comparison. Even  
18 during this time declines in control hive condition are commonly  
19 encountered.

20 Furthermore, effects at other critical time  
21 periods of hive development, such as overwintering, are not  
22 addressed. Although such effects could be assessed if the post  
23 tunnel observation period were extended and colonies were of  
24 sufficient strength to support overwintering.

25 For the feeding studies, the bees are allowed  
26 to freely forage, so consumption of the spiked food provided to



1 the colony depends on a range of factors that may not be  
2 constant for all experimental conditions. For example, if  
3 alternative forage is ample, then the bees may use less spiked  
4 sucrose solution or spiked pollen then when outside forage  
5 sources are scarce. Thus, this type of study, which does not  
6 use an enclosure, is subject to numerous sources of variability  
7 at the field level, including weather, disease, parasites and  
8 exposure to other pesticides present in the environment.

9 On the second bullet, the feeding aspect is  
10 artificial and may not accurately mimic typical foraging  
11 behavior by honey bees, or the stability of the test material in  
12 such matrices under natural conditions.

13 The third bullet, the foragers only need to  
14 move minimal distances to the food source, therefore, use of an  
15 internal spiked food source can confound efforts to integrate  
16 impacts on foraging behavior when foragers feed on a treated  
17 crop.

18 The fourth bullet, the performance of the  
19 colonies has not been evaluated in relation to long-term  
20 continual use of the artificial food source. And the final  
21 bullet notes that as a study increases in duration, it also  
22 increases in complexity and potentially confounding factors,  
23 such as the interventions required to enable a colony to  
24 overwinter.

25 It is essential to highlight the importance of  
26 establishing the statistical significance necessary to



adequately evaluate a desired level of protection based on biological significance. The issue of statistical and biological significance relates to both semi-field and full field studies; the latter of which will be explored later in the presentation.

Both of these types of studies seek to evaluate the potential for an effect, relative to the control group at a specified application rate or concentration within a supplied food source. In some cases, we are able to evaluate time trends and hive performance, though these are not routinely reported. Yet there is a high level of variability typically associated with the measurement endpoints in both types of studies, where variability may be so high that it is difficult to isolate any effect.

While 10 percent mortality has been established as a level of protection at the screening level, a level of biological significance has not yet been established that links measurement endpoints at either the semi-field or the full field level to the assessment endpoints. Furthermore, because of compensatory mechanisms within the colony, apparent effects may only be transitory.

Well designed semi-field studies, despite their uncertainties, should be able to inform the risk manager by providing clear lines of evidence linking multiple routes of exposure to adverse ecological effects and provide for a more robust characterization of the risk related to the assessment



1 endpoints.

2           Semi-field studies assess both exposure and  
3 effects at the colony level that are less conservative and more  
4 realistic than laboratory assays on the individual bees. These  
5 studies also integrate effects from toxic degradates, sublethal  
6 effects and chronic exposure.

7           Furthermore, the Tier II feeding study for  
8 adults and brood should provide a no effect concentration that  
9 can then be compared to residue levels in various crops  
10 identified in targeted residue studies.

11           Unlike the Tier I toxicity studies that are  
12 used to calculate risk quotients, the use of the Tier II studies  
13 will be used to qualitatively evaluate risk and to help  
14 characterize the quantitative risk estimates derived from  
15 laboratory studies on the individual at the first tier.

16           Consequently, semi-field studies will help  
17 inform the risk manager regarding any need for mitigation  
18 options or options for further refinement. When uncertainties  
19 remain from the Tier II assessment or risk cannot be mitigated,  
20 the risk manager may decide that additional data are necessary  
21 in the form of Tier III full field studies.

22           The transition to Tier III is based on  
23 addressing remaining uncertainties at the previous tiers. This  
24 slide highlights some of the major uncertainties inherent to the  
25 semi-field study designs and how the Tier III full field study  
26 may address those design limitations. In the case of the tunnel



1 study, a full field study does not employ tunnels and therefore  
2 eliminates the stress of the tunnel on the various aspects of  
3 colony functioning, as well as limitations in the size of the  
4 colonies and the duration of the study.

5 Related to the feeding designs, a Tier III  
6 full field study allows hives to naturally forage and it also  
7 allows a study to integrate any impacts on the foragers that  
8 visit the treated crop or consequences to the hive based on  
9 foraging on the treated crop.

10 In summary, the full field study overcomes the  
11 major limitations of the stress placed on the colonies from the  
12 enclosure in the tunnel design and the artificial exposure  
13 pathway in the feeding designs.

14 A Tier III study should further refine any  
15 uncertainties identified by the Tier II assessment, related to  
16 the assessment endpoints. The Tier III study is a full field  
17 design that provides the highest level of refinement of the  
18 tunnel and feeding study designs.

19 Consequently, Tier III effects assessments are  
20 intended to represent highly refined studies that address  
21 specific uncertainties and/or risks identified in lower tiered  
22 studies.

23 Similar to the transition from Tier I to Tier  
24 II, the risk manager will consider the availability of multiple  
25 sources of information. The need to transition to Tier III is  
26 therefore not an automatic requirement, but rather, based on



1 multiple lines of evidence.

2 A field study serves as a means of addressing  
3 uncertainties raised in lower tier studies conducted either in  
4 the laboratory or under semi-field conditions and can be useful  
5 in examining the effects of pesticides with extended residual  
6 toxicity.

7 However, a full field study is intended to  
8 represent realistic application conditions, whereby the test  
9 substance is applied to a specific crop in which bees are  
10 foraging freely, without the use of an enclosure, and whereby  
11 test colonies are primarily exposed through residues carry back  
12 to the hive by the forager bees.

13 Consequently, a full field study is also the  
14 most resource-intensive, relative to studies used for the Tier I  
15 and II assessments, to conduct in terms of both time and money.  
16 These studies can also be the most difficult to interpret given  
17 the level of associated variability. Therefore, they can  
18 represent a significant investment to regulatory agencies in  
19 terms of the resources that are required to review these  
20 studies, but these studies may also provide contacts relative to  
21 other lines of evidence.

22 By serving as a study to refine the  
23 uncertainties and effects identified in the previous tiers, the  
24 field study is also meant to test for the potential of an effect  
25 and not the mechanistic cause of an effect. Furthermore, the  
26 Tier III field study should have a focused risk hypothesis, that



1 is, a risk hypothesis informed by and built upon the previous  
2 tiers.

3 There are a number of guidance documents  
4 related to the conduct of a field study. Currently, the EPA has  
5 guidance for conducting the pollinator field study.

6 Similarly, the EPPO Guidance also addresses  
7 pollinator field study design. Regarding the first major  
8 bullet, the EPA field pollinator guidance does not provide much  
9 detail, but rather, provides flexibility for designing a study  
10 based on the needs of the risk assessment.

11 The second major bullet mentions Guideline  
12 850.2500. Although intended as guidance for conducting field  
13 studies with mammals and birds, this EPA Guidance titled, "Field  
14 Testing for Terrestrial Wildlife" provides very useful  
15 information on study design elements to consider for field  
16 testing.

17 This guidance addresses the basic purpose of a  
18 field study, geographic area and selection of study sites,  
19 sample size, and the test conditions of the study. EPPO  
20 Guidance provides more specific detail that can be used to  
21 design a field pollinator study with honey bees in terms of  
22 experimental conditions, such as selection of the crop,  
23 placement of the colonies, relative to the treated field, size  
24 of the colonies and plot size.

25 Also, application of treatments, such as use  
26 of controls, timing of application and rates; and finally, mode



1 of assessment, such as timing and frequency of assessments.

2           Similar to Tier II studies, full field studies  
3 are flexible in terms of their specific design, as they are  
4 based on uncertainties highlighted in the previous tiers, as  
5 well as the nature of the pesticide. Consequently, registrants  
6 should submit protocols for the field study prior to study  
7 initiation in order to identify the key design elements based on  
8 specific risk management concerns. In general, though, there  
9 are design elements common to most field studies.

10           The second bullet notes that a field study for  
11 honey bees, in general, should be conducted under actual use  
12 conditions in which a typical end-use product is used at its  
13 maximum application rate, frequency and method, as described in  
14 the label for a specific crop. The design should reflect a  
15 realistic foraging environment and exposure system in the field  
16 where the pesticide is to be applied.

17           The third bullet notes that the study is  
18 conducted in a crop grown outdoors without enclosures.  
19 Consequently, the primary exposure route is to foragers that are  
20 allowed to freely fly and forage.

21           The fourth bullet notes that the crop is  
22 subject to good agricultural practices. And the fifth bullet  
23 mentions the size of the hives, each with a minimum of  
24 approximately 10,000 bees which are placed within or at the edge  
25 of treated or controlled flowering plots and monitored for a  
26 specified period of time, depending on the concerns of the



1 chemical, as in the case of a systemic compound that is  
2 persistent in high food stores.

3 Finally, the last bullet deals with the  
4 duration of the study. The evaluation of the hive should  
5 ideally proceed for a minimum of 50 days, based on the lifecycle  
6 of the honey bee within the colony. Because hives may differ in  
7 size, colonies should be distributed as equitably as possible  
8 between treatments. The colonies should be in position  
9 approximately two to three days before the trial in order to  
10 acclimate to the study site.

11 If the chemical is a foliar spray, treatments  
12 should be applied when the test crop is in full flower during  
13 the daytime when bees are demonstrated to be actively foraging  
14 on the test crop. Hives should be queen ripe; that is to say  
15 have a single functional queen. Use sister queens that is a  
16 common maternal source and be of similar strength at the start  
17 of the study.

18 Equipment should be free of other chemicals  
19 and the hives should have low incidences of disease and  
20 parasites, though it is nearly impossible to use hives complete  
21 free of disease and pests. Unlike Tier II studies, there are  
22 currently no pesticides identified as an adequate positive  
23 control at the full field Tier III level.

24 The range of endpoints that may be measured,  
25 as well as the selection of endpoints in the Tier III full field  
26 study are similar to those in the Tier II semi-field study. As



1 mentioned in the Tier II section, the endpoints should also be  
2 specific to the uncertainties identified earlier in the tier  
3 process as they relate to the assessment endpoint so as to  
4 minimize the number of endpoints to be collected and the  
5 continued opening of the hives.

6 Also similar to the Tier II study designs,  
7 Tier III studies integrate toxic degradates, chronic exposure  
8 and sublethal effects into the measurement endpoints. A major  
9 advantage of the Tier III field study is that the duration of  
10 the study with possible overwintering and the consequent timing  
11 of measurements are flexible and could be suited to the  
12 properties of the chemical under actual use conditions.

13 In order to adequately interpret a field  
14 study, it would be helpful for the study to properly evaluate  
15 exposure. It can be difficult to evaluate the effects of the  
16 test chemical within a full field study because the design  
17 allows for bees to forage freely and to obtain food both onsite  
18 and off the treated site.

19 In studies with limited bloom density on the  
20 treated crop and extensive alternative forage in the vicinity,  
21 bees may actually forage very little in the test plot and this  
22 may lead to an underestimation of potential effects on the hive  
23 due to reduced exposure or alternatively to an inability to  
24 detect treatment effects due to controlled contamination

25 There are a number of ways of measuring the  
26 degree to which the colony is exposed to the test substance.



1 First, pollen traps are fixed at the entrances of the hives,  
2 force pollen off of the legs of a percentage of foragers when  
3 they return to the hive. This pollen can then be analyzed for  
4 residues.

5 Second, freshly stored nectar and pollen or  
6 stored bee bread and honey can be measured in the hives to  
7 determine the level of residues that the bees are storing for  
8 consumption or processing prior to feeding of the queen and  
9 brood.

10 Measures of residues in the blossoms would  
11 provide a comparison of stored concentrations with those  
12 collected from the blossoms. When pollen is collected from the  
13 pollen trap, the study may analyze floral origin and percent  
14 composition of the pollen. Some pollen from the flowers may  
15 also be collected by the bees and mixed with the nectar when it  
16 is stored. So in addition to sampling of residues and freshly  
17 stored nectar, pollen could potentially be obtained from within  
18 this collected nectar to determine the floral origin of the  
19 nectar as well.

20 Finally, qualitative measures of foraging  
21 activity, potential quantitative measures of the return of  
22 forager bees and quantitative measure of bloom density can  
23 provide information on the potential attractiveness of the crop  
24 of interest.

25 The Tier III full field study is complex and  
26 difficult to carry out. So there are a number of design



1 challenges that we face and need to address. Consequently, we  
2 are requesting feedback on the design challenges of the field  
3 studies and our suggestions for improving these designs that  
4 will be covered later in the presentation.

5           Concerning the design challenges, honey bees  
6 have an extensive foraging range. Given an alternative to  
7 forage, honey bees may minimally forage on the test fields as  
8 there is no means of controlling where honey bees forage. In  
9 addition, uncontaminated food reserves collected prior to study  
10 initiation may result in delayed exposure of the colonies to the  
11 test substance, as these previously collected food reserves may  
12 be used first.

13           The weather can also affect the ability of  
14 bees to forage, where similar to the Tier II studies, adverse  
15 weather may prevent honey bees from foraging on a test crop.  
16 Finally, to date, typical plot sizes in field studies are  
17 minimal, relative to both the representative acreage of some  
18 crops in North America and the potential foraging radius of the  
19 honey bee.

20           This slide represents the range of plot sizes  
21 for typical field studies ranging in sizes from two and a half  
22 to 13 acres, which equates to .004 to .02 square miles.  
23 Relative to the maximum foraging area of the honey bee colony,  
24 based on a five-mile foraging radius, this represents .0051 to  
25 .025 percent of the potential maximum foraging area. The  
26 question then remains: what size a plot would satisfy the needs



1 of the colony to ensure that it forages to the maximum extent on  
2 the treated field; especially considering that replication of  
3 the colonies at the within plot level is typically three to four  
4 full size hives that can each be up to 50,000 to 60,000 bees.

5 The colony size is another factor to consider.

6 The EFSA Scientific Opinion and pollinator risk assessment also  
7 notes several points related to the advantages of different  
8 sizes of the hives for the field study. Larger populations may  
9 be more efficient honey producers and produce more honey on a  
10 per bee basis while using less per bee over the winter.

11 Thus, a larger colony with more foragers and  
12 in nest bees could provide a better observation of the effects  
13 of the pesticide because of the higher number of bees involved.  
14 However, the EFSA Scientific Opinion also notes that smaller  
15 colonies may be more sensitive due to reduced resilience to  
16 replace foragers with nurse bees in a smaller brood area.

17 The second major bullet notes that hives at  
18 the full field level cannot be completely protected from the use  
19 of other pesticides, either on the test field itself or adjacent  
20 to the test fields. These products may be applied to the same  
21 crop as part of grower's standard practice or they may be  
22 applied on neighboring fields on which honey bees may forage or  
23 from which the pesticides may drift into the test or control  
24 plots of a full field study.

25 In the case of persistent pesticides, these  
26 chemicals may remain on the test field or in the adjacent field



1 well after the application. These products may then be  
2 collected by honey bees and brought back to the hive, thereby  
3 serving as a source of exposure to other chemicals or even the  
4 chemical under study.

5 As the third major bullet highlights, the  
6 treatment and control fields should also be adequately  
7 separated, though logistical constraints associated with  
8 replication of sites may make this difficult.

9 For control in treatment plots with inadequate  
10 separation where honey bees can forage freely, bees from control  
11 plot may forage on treated plots, thereby bring contaminated  
12 food into the control hives. Conversely, treatment plot hive  
13 bees may forage in control plots as well, thereby decreasing the  
14 extent of exposure. This movement of control and treatment plot  
15 bees between plots can confound the interpretation of the field  
16 study.

17 Parasites and certain diseases can also affect  
18 honey bee colonies, as the fourth major bullet states. Some of  
19 these can impact the same measurement endpoints as the pesticide  
20 of interest.

21 The next major bullet highlights the duration  
22 of the study that also presents a challenge, especially when the  
23 study is carried through overwintering and attempts to ascertain  
24 the effect of a pesticide while minimizing any potential  
25 confounding effects of the interventions that are required to  
26 enable a honey bee colony to survive overwintering.



1           The final bullet addresses replication and  
2     variability. As previously noted in the section on Tier II  
3     studies related to variability and biological significance, the  
4     level of variability in the measurement endpoints is relatively  
5     high, due to all of the potential confounding factors at the  
6     full field level.

7           As a result of these design challenges, we  
8     have identified a number of modifications that may enable more  
9     robust field designs. The size of the plots should increase to  
10    be more reflective of the size of representative crop acreage in  
11    North America. The size of the areas should also adequately  
12    support the nutritional requirements of the hive for the  
13    duration of the study with the specific purpose of maximizing  
14    the foraging of the hives on the test crop to provide an upper  
15    bound exposure scenario for that crop.

16          It is recognized, however, that the bloom  
17    period of the test crop may be short, relative to the overall  
18    study period. But the study design must consider that the  
19    nutritional requirements of the study colonies are met.

20          A field study should also provide for adequate  
21    separation of treated and control plots, however, climatic and  
22    landscape variations may preclude greater distances between  
23    plots, depending on these variations over varying geographic  
24    scales.

25          According to the EFSA Scientific Opinion, the  
26    field test sites should use areas with similar environmental



1 conditions, where possible, with at least four to six kilometers  
2 that is 2.5 to 3.7 miles between treated and control plots.

3 Statistical analysis should be conducted for  
4 all field studies; therefore, field studies should also  
5 determine the power of the test to adequately assess the  
6 endpoints relative to the appropriate level of biological  
7 significance. An appropriate power analysis should accompany  
8 each protocol proposal in order to determine the number of hives  
9 necessary for the conduct of the study.

10 In addition, the final report should also  
11 contain an appropriate power analysis to determine if the level  
12 of statistical power has been achieved. Measures of exposure  
13 are essential to the interpretation of the field study.  
14 Consequently, it would be helpful for the study to measure  
15 residues in incoming pollen through the use of pollen traps  
16 and/or nectar, as in sampling of the honey bee stomach; measure  
17 residues and store pollen and nectar pre-exposure and at various  
18 times, post-exposure; measure bloom intensity and duration of  
19 that intensity and identify sources and percent composition of  
20 pollen and nectar.

21 The studies should also limit disease and  
22 pests using new hive equipment and obtain bees from sources with  
23 low pest occurrence. This slide presents other modifications  
24 that could help to improve the quality of the field studies that  
25 regulatory agencies receive. The duration of the study should  
26 match the concern of the chemical. According to the EFSA



1 Scientific Opinion, the colonies that are used in the experiment  
2 should be monitored for a period of time, covering the entire  
3 flowering period and last at least two brood cycles. When  
4 residues are persistent, monitoring should continue through  
5 winter to after the overwintering period.

6 Other recommendations proposed by the EFSA  
7 Scientific Opinion, related to field study designs, include the  
8 use of a highly attractive crop, quantitative assessment of  
9 brood condition, residue analysis of in hive bees and evaluation  
10 of the storage of food and their associated residue levels, and  
11 in some cases, the removal of food stocks from the hive at study  
12 initiation to maximize the exposure to the test substance.

13 Given the previously mentioned modifications  
14 to the design of the field study, it is important to note that  
15 some of them notably increasing the plot sizes and ensuring  
16 adequate replication would likely be difficult to implement,  
17 given the logistics of a field study.

18 The purpose of these studies is that they  
19 provide important information relative to the risk assessment.  
20 Note, that as we move up the tiers to Tier III, we not only  
21 increase the realism, but we also increase the variability due  
22 to the number of potentially confounding factors as well as the  
23 cost, in terms of time, money, and resources.

24 In fact, full field studies have cost a  
25 considerable amount of resources, both to conduct and review,  
26 without being classified as acceptable, and only providing



1 minimal gains, in terms of the risk assessment.

2 In these cases, field studies have not been  
3 particularly refined, but rather, measured numerous endpoints,  
4 whether quantitatively related to the assessment endpoints  
5 outlined earlier or not.

6 Furthermore, there is uncertainty in the  
7 results, due to factors, such as minimal distance between  
8 treated and control plots that allows the forager bees of the  
9 two treatment groups to cross forage; the exposure of test  
10 colonies to numerous other chemicals that are used as standard  
11 grower practice and the lack of adequate replication given high  
12 levels of variability and the measurement endpoints.

13 To date, for the studies that we've seen  
14 related to the exposure studies, the refined exposure studies,  
15 there appears to be less variability in those studies relative  
16 to the ones that assess effects to the bees at the whole hive  
17 level.

18 The previously mentioned modifications to the  
19 Tier III field study should enable a stronger, more robust  
20 design. A properly design and implanted field study should  
21 therefore provide essential information on actual uses of the  
22 pesticide of interest that can inform the risk manager in any  
23 additional mitigation options, if available.

24 The field study should also provide a more  
25 clear linkage between the measurement endpoints and the  
26 assessment endpoints from the actual use of the chemical, as a



1 field study can be screening or definitive in nature to either  
2 determine the presence or absence of an effect or define the  
3 magnitude and duration of an effect.

4 The ultimate design of the study will be  
5 dictated by the specific uncertainties it is intended to address  
6 in order to qualitatively describe the risk, effects and  
7 uncertainties identified in the lower tiers. While these  
8 studies are conducted in response to concerns identified in  
9 lower tier studies, the full field study is ultimately intended  
10 to address whether risks estimated using lower tier exposure and  
11 effect data occur under actual use conditions.

12 It is also important to note that the field  
13 study will not be interpreted in isolation of the other tiers  
14 and other information. Rather, we will evaluate and interpret  
15 the field study and their risk assessment using all of the data  
16 available to the risk assessor, including data from laboratory  
17 toxicity studies at the first year, open literature, non-target  
18 arthropod studies, sublethal effects, incident reports, other  
19 semi-field or full field studies and the residue levels in  
20 various crops, based on exposure data. This weight of evidence  
21 approach will be further explained in the next presentation.

22 Thank you for your time. We can now take any  
23 questions that the panel may have.

24 **DR. DANIEL SCHLENK:** Okay. Dr. Berenbaum.

25 **DR. MAY BERENBAUM:** I may have missed this,  
26 but in transitioning from Tier I to Tier II to Tier III, is



1 there a task that assumption that colony genetics will be  
2 controlled for or minimally subspecies? Is there a  
3 consideration of consistency in terms of genetic identity from  
4 one tier to the next?

5 **DR. THOMAS STEEGER:** At this time, we're  
6 not proposing the use of doing genetic analyses to ensure that  
7 the bee populations that are typically used in these studies are  
8 drawn from healthy hives and presumably reflect a relatively  
9 broad genetic base.

10 **DR. DANIEL SCHLENK:** Dr. Hunt.

11 **DR. GREG HUNT:** I'm wondering, have there been  
12 any full field studies that have been classified as acceptable  
13 by the EPA?

14 **DR. THOMAS STEEGER:** We have classified  
15 studies as acceptable as having met the standard or the  
16 guideline requirement for conducting a full field study, but as  
17 was pointed out by Joe DeCant, the 850.3040 guideline is  
18 relatively broad and does not specify specific expectations that  
19 would have to be met or specific criteria that would have to be  
20 met to downgrade a study unless things that have happened, such  
21 as contamination, have reached a point that the ability of the  
22 study to differentiate a treatment level effect has been  
23 completely compromised.

24 If the study is scientifically unsound it will  
25 be downgraded, but we have accepted field studies; but primarily  
26 because there aren't strong criteria for potentially downgrading



1 those, except best professional judgment that the scientific  
2 integrity has been compromised.

3 **DR. GREG HUNT:** Thank you. That answers my  
4 question. I was just concerned about the level of cost and time  
5 in doing these studies.

6 **DR. DANIEL SCHLENK:** Mr. Pistorius.

7 **MR. JENS PISTORIUS:** Would it be possible to  
8 skip Tier II and go to Tier III directly?

9 **DR. THOMAS STEEGER:** Again, the transition  
10 from screening level assessments to higher tier assessments are  
11 a risk management decision. It is entirely possible to  
12 transition from a screening level to a full field study;  
13 however, the intent, as you move up in refinement, is to refine  
14 the uncertainties; and I'm not sure that you would have been  
15 able to do that by transitioning quickly to a full field study  
16 from what could be gleaned from laboratory studies, unless you  
17 had available to you open literature studies or incident data  
18 that would enable you to do those types of refinements so that  
19 you can better fine tune the risk hypothesis that's being tested  
20 at the full field level.

21 **MR. JENS PISTORIUS:** Another one. What would  
22 be the basis for election of which Tier II test has to be  
23 conducted, taking into account the different limitations of the  
24 described methods?

25 When would you say use this test, use this  
26 test, free feeding, Oomen, et al. and all these different kind



1 of --

2 **DR. THOMAS STEEGER:** I think that you're  
3 asking some very excellent questions and we're hoping that the  
4 SAP can provide insight on that. Again, the planning dialogue  
5 that takes place as part of the risk assessment process -- the  
6 risk assessor will inform the risk manager of what kind of  
7 uncertainties there are and we attempt to work with the  
8 regulated community in terms of how best to address those  
9 uncertainties.

10 So whether you use a semi-field tunnel study  
11 or whether you use a feeding study or you use the Oomen method,  
12 those are all on the table for discussion in terms of what  
13 direction the Agency will actually require the registrant to  
14 take.

15 **DR. DANIEL SCHLENK:** Okay. Dr. Fefferman.

16 **DR. NINA FEFFERMAN:** Forgive me if this is a  
17 really silly question, it relies on two things I don't know of  
18 honey bee physiology and basic toxicology.

19 The discussions I've been hearing about  
20 toxicity levels, again, I know you're mainly focusing on the  
21 acute side of things, but on the chronic side, the flip side of  
22 exposure, of course, is elimination.

23 I was wondering whether or not these are  
24 things such that once they're ingested they stay in the bee  
25 permanently or if there is a rate of elimination kind of  
26 question that we should be asking. And if so, if there's any



1 room in the kind of toxicity studies that you're running to look  
2 at the durational dose response curves and dose maintenance  
3 curves in a bee over its individual life and then possibly life  
4 of a colony.

5 I'm thinking specifically of things that might  
6 be pulse treatment types of pesticides and whether or not that  
7 will actually be a constant daily level or whether or not as  
8 long as they survive the main pulse and we don't pulse them  
9 again until that's out of their system that might be okay. That  
10 kind of thing.

11 **DR. THOMAS STEEGER:** I think you raised  
12 some very interesting questions. The screening level assessment  
13 is obviously testing or focused on acute toxicity and the  
14 exposure estimates that we're using are based on the presumption  
15 that none of the compound is degrading, so that's why they're  
16 very conservative assessment estimates. But as you move up in  
17 higher levels of refinement and the duration of these studies is  
18 more protracted, presumably you are able to incorporate how the  
19 chemical is moving through the colony, through the animal and  
20 looking at its potential to rebound from the effects that  
21 might've been observed at lower level studies.

22 So actually attempting to isolate out the  
23 formation and decline curves of compounds within the bee can be  
24 a problem because you're going to have to be manipulating the  
25 colony quite a bit. And as Joe DeCant had indicated, the more  
26 that you manipulate the colony the more variability it's likely



1 that you're going to introduce into it.

2 So our focus at this time is trying to collect  
3 data points that provide us a sense for the colony level  
4 response, as opposed to the individual bee response and under as  
5 natural conditions as possible without over manipulating the  
6 colonies to try to get at those answers.

7 **DR. DANIEL SCHLENK:** Do you have something  
8 to add?

9 **MR. KEITH SAPPINGTON:** I just wanted to add  
10 that some of the characteristics of the compound -- while I  
11 agree in the Tier II and Tier III level you would have a  
12 long-term exposure -- but some of the characteristics of the  
13 compound, particularly its persistence in bioaccumulative  
14 abilities or characteristics would be sort of indicators, up  
15 front, that you might consider longer term accumulation within  
16 the organism. We held a separate SAP about four years ago on  
17 this whole process of how you do risk assessment for persistent  
18 bioaccumulative and toxic chemicals.

19 With that said, we have a process for aquatic  
20 organisms that relies on bioaccumulation modeling as well as  
21 measured information in bioaccumulation tests. And there are  
22 some models that we use as a kind of a screen, up front, in  
23 problem formulation for terrestrial organisms. But to my  
24 knowledge, I have not seen a model yet that looks specifically  
25 at the toxicokinetics and accumulation within the bee; but at  
26 least we might get an indication early on.



1 DR. DANIEL SCHLENK: I think Dr. Potter  
2 had a --

3 DR. THOMAS STEEGER: This is Tom Steeger.  
4 I'm sorry.

5 DR. DANIEL SCHLENK: Go ahead. One more.

6 DR. THOMAS STEEGER: I'd just like to add  
7 onto a question that had been asked earlier by Dr. Potter  
8 regarding the utility of the higher tier field studies. My  
9 interpretation of your question was relative to full hive  
10 studies.

11 If your question was relative to exposure,  
12 higher tier exposure studies, those studies have proven to be  
13 very effective means of documenting the level of exposure that  
14 colonies or individual bees are exposed to.

15 We've been working collaboratively with the  
16 California Department of Pesticide Regulation and with Health  
17 Canada Pest Management Regulatory Agency to collect those  
18 studies from field-based information and they are very useful  
19 for documenting exposure.

20 DR. DANIEL SCHLENK: Dr. Potter.

21 DR. THOMAS POTTER: I guess as a follow-up,  
22 are information from those studies imbedded in this risk  
23 assessment process that we're looking at here today?

24 DR. THOMAS STEEGER: We are proposing that  
25 was depicted in the flow chart that Keith Sappington discussed  
26 and as Chris Garber discussed as well in her presentation that



1 -- it's not up on the board -- but Box 6(a) is where you're  
2 evaluating potential exposure through RQ values, through  
3 field-based studies and those estimates of exposure that are  
4 actually measured values feed back into the calculation of RQ  
5 values that would hopefully, potentially reduce the RQ value  
6 below the level of concern.

7 So what is inherent in this risk assessment  
8 framework that we are proposing is that field level measurement,  
9 whether they're collected either from the semi-field environment  
10 or from a full field study where you're actually focusing on  
11 exposure, you're measuring residues in pollen and nectar, would  
12 then be used at the numerator in the risk quotient that we'll be  
13 talking about in the next section that would be able to  
14 potentially -- at a screening level, where you're still  
15 quantifying risk -- to get risk quotients below LOCs and be able  
16 to come to a presumption of minimal risk.

17 **DR. THOMAS POTTER:** Can I follow up on that?

18 **DR. DANIEL SCHLENK:** Sure.

19 **DR. THOMAS POTTER:** In looking through the  
20 White Paper, I was struck by what I perceived to be a lack of  
21 information or good quality data regarding concentrations of  
22 active ingredients in pollen and nectar so that to a large  
23 degree, we're dependent on a limited number of studies to  
24 compare the proposed exposure levels to.

25 If there is additional information that's been  
26 submitted as part of the registration process, it certainly



1 would seem to me to be very helpful, in terms of supporting this  
2 entire process to somehow get that data out to the light of day.

3 **DR. THOMAS STEEGER:** I think that Dr.  
4 Berenbaum raised that same issue in terms of what appears to be  
5 relatively dated information. Those are published studies, but  
6 the Agency does have registrant-submitted studies that have been  
7 conducted either on the initiative of the regulated community or  
8 in response to data call-ins, such as that of California's, that  
9 are providing that very information and we deem it to be high  
10 quality information that would enable us to test the  
11 assumptions.

12 In fact, we've compared some of these measured  
13 residues against the model estimates, the Chemical X or Chemical  
14 1, the undisclosed chemical. That information is still  
15 considered confidential business information, but those are  
16 actually measured values that some of these model estimates and  
17 default values are being compared to.

18 **MS. KRISTINA GARBER:** Just to add a little bit  
19 more, we did actually look at all the registrant-submitted data  
20 that we had for pollen and nectar and we incorporated everything  
21 we could in the White Paper.

22 **DR. THOMAS POTTER:** I will note that if you  
23 look at the executive summary of the Pellston conference -- I  
24 believe it's in the European Risk Assessment document as well --  
25 that identified the need for data that can be used to support  
26 the risk assessment process as a critical need. So I'll leave



1 it at there. We can go around in circles with that, but thank  
2 you.

3 **DR. DANIEL SCHLENK:** Any other questions  
4 related to the presentation?

5 (No response.)

6 Okay. Dr. Steeger, do you want to move us  
7 ahead?

8 **DR. THOMAS STEEGER:** Thank you for this  
9 opportunity to address the Scientific Advisory Panel. Again, my  
10 name is Tom Steeger. I'm a biologist and I'm a senior science  
11 advisor in the Environmental Fate and Effects Division.

12 In this presentation, I will discuss the risk  
13 characterization component of the risk assessment process and  
14 how risk characterization integrates exposure and effect  
15 estimates with other lines of evidence to provide risk managers  
16 with an understanding of potential risk from the proposed use of  
17 the chemical.

18 I'll also discuss how the risk  
19 characterization articulates the assumptions, limitations, and  
20 uncertainties associated with the available data.

21 The risk characterization is intended to  
22 provide risk managers with an overall understanding of potential  
23 risks to the environment. In doing so, the assessment must be  
24 transparent, that is, it must be explicit with respect to the  
25 process that was followed. The risk assessment must be clear  
26 and easy to understand and consistent with Agency guidance and



1 reasonable such that the risk manager and any stakeholder can  
2 understand how conclusions were arrived at, such that the risk  
3 assessment could be repeated and reach a relatively similar  
4 conclusion given the same process, underlying data and  
5 assumptions.

6 As was discussed in the preceding  
7 presentations by Tom Moriarty and Keith Sappington this morning,  
8 the risk assessment process consists of three phases or steps:  
9 the problem formulation phase, the analysis phase, where effects  
10 are characterized and exposure is characterized, and that's  
11 followed by the risk characterization phase.

12 Although the flow chart on the left is  
13 depicted as unidirectional, as Keith had pointed out earlier,  
14 the various phases in the risk assessment are intended to be  
15 iterative and as more information becomes available through the  
16 analysis phase, the problem formation may evolve.

17 Communication with risk manager is an  
18 essential component of the risk assessment process since  
19 potential mitigation measures or other additional data can be  
20 evaluated in the context of how exposure estimates may be  
21 affected.

22 Once the analysis phase is concluded, the risk  
23 assessment process proceeds to the risk characterization phase.  
24 This phase consists of two components: the risk estimation and  
25 the risk description.

26 During risk estimation step of risk



1 characterization, risk estimates are calculated. Risk estimates  
2 are based on a deterministic approach using point estimates or  
3 providing that there are sufficient data, estimates can be based  
4 on a distributional approach.

5 Although not discussed extensively in the  
6 White Paper, distribution-based approaches, as opposed to point  
7 estimate-based approaches, can be use to characterize both  
8 exposure and effects and joint probability curves constructed to  
9 then estimate the likelihood and magnitude of an adverse effect.

10 However, in today's presentation, we are  
11 focused on point estimate-based approach, which compares risk  
12 estimates to levels of concern and the potential for risk as  
13 articulated in terms of whether the level of concern is exceeded  
14 or not.

15 The proposed framework for estimating risk at  
16 a screening level is consistent with the process currently used  
17 by OPP to estimate risk to other taxa, and is based on the  
18 concept of risk being a function of both exposure and toxicity.  
19 Point estimate of environmental concentrations, such as those  
20 described earlier by Kris Garber, are compared to point  
21 estimates of toxicity endpoints discussed by Joe DeCant.

22 The quotient of the ratio of exposure to  
23 effects is the risk quotient. As discussed by Kris Garber, at  
24 the screening level, point estimates of exposure are initially  
25 based on relatively conservative models or default values.

26 However the risk assessment process is



1 intended to be iterative, and as discussed by Reuben Baris,  
2 exposure values used to generate RQs can be further refined  
3 through the use of measured values obtained through other  
4 studies, typically submitted to EPA, such as magnitude of  
5 residue studies or through targeted monitoring studies of  
6 residues and foliage or in pollen and nectar.

7 Table 22 of the White Paper provides a summary  
8 of the proposed exposure and effect point estimates used to  
9 calculate risk quotients and the screening level risk assessment  
10 for pesticides applied foliarly, as seed treatments, soil  
11 treatments, and tree trunk injections.

12 This is an excerpt from the table and it's  
13 taken from Table 22, for pesticides applied to seed treatments,  
14 the dietary-based RQ values for adults is based on the products  
15 of the Briggs model estimated environmental concentration,  
16 multiplied by .29 grams per day, serving as a numerator, divided  
17 by the acute oral LD50 value for individual adult bees.

18 RQ values estimated in the risk estimation  
19 phase of the risk characterization are compared to the Agency's  
20 level of concern. Levels of concern represent the Agency's  
21 interpretive policy and are used to analyze potential risk to  
22 non-target organisms and the need to consider regulatory action.

23  
24 These criteria are used to indicate when a  
25 pesticide is used as directed on the label, has the potential to  
26 cause adverse effects on the non-target organisms.



1 Historically, the Environmental Fate and Effects Division has  
2 used the default probit dose response slope of 4.5 to estimate  
3 acute effects to organisms; however, the level of concern for  
4 estimating acute risk to honey bees is based on an analysis of  
5 honey bee acute contact and acute oral toxicity studies that had  
6 been submitted to EPA.

7 This analysis resulted in a mean probit dose  
8 response slope for acute contact toxicity at 3.93 and a mean  
9 probit dose response slope for acute oral toxicity studies of  
10 3.4. The Environmental Fate and Effects Division has relied on  
11 the Individual Effect Chance model, which is abbreviated the  
12 IEC, to estimate the likelihood of an effect, for example, acute  
13 mortality.

14 This model uses the calculated risk quotient  
15 and the probit dose response slope that integrates those values  
16 to derive the probability of an individual mortality. However,  
17 the model can be used in reverse to estimate the risk quotient  
18 value that would be needed to result in a particular magnitude  
19 of an effect, given a specific dose response slope.

20 Since data are available for both acute oral  
21 and acute contact lethality studies, both sets of data were  
22 combined. Rather than using the mean dose response values,  
23 though, the median dose response slope across all acute contact  
24 and oral toxicity studies was used as a measure of central  
25 tendency and this median dose response slope is 3.2.

26 Using the individual effect calculated model,



1 the level of concern was then back calculated, which was a  
2 result in 10 percent mortality based on the median dose response  
3 slope of 3.2. This slide depicts Table 24 from the White Paper,  
4 summarizing that the median slope of 3.2 and the likelihood of  
5 10 percent mortality that is in effect, the LOC of 0.4 is  
6 necessary.

7 The 10 percent level of effect, that is the  
8 acute mortality, is based on the maximum level of mortality  
9 permitted in controls. That is, untreated studies of acute oral  
10 and contact toxicity based on current guideline recommendations.

11 Although the chronic toxicity study guidelines  
12 are not currently available for honey bees the proposed chronic  
13 risk level of concern is 1) since the RQ value for estimating  
14 chronic risk is the ratio of the maximum estimated exposure  
15 concentration to the no-observed adverse effect concentration;  
16 that is, the NOAEC, an LOC of one is consistent with that used  
17 for evaluating chronic risk to other taxa.

18 As discussed by Mr. Sappington and as depicted  
19 in Figures 2 and 3 of the White Paper showing the proposed risk  
20 assessment framework for honey bees, the process is intended to  
21 be iterative. The slide depicts the risk estimation portion of  
22 the framework where risk quotients are calculated in Boxes 4(a)  
23 and 4(b), based on conservative exposure estimates derived in a  
24 screening level assessment.

25 Charge Question 13 asks the SAP to provide  
26 comment on the exposure and effect data used to estimate risk



1 and the acute and chronic risk LOCs that are in turn used to  
2 evaluate the risk quotients. Question 13 also asks the SAP to  
3 comment on how other line of evidence are integrated into the  
4 risk assessment during the second phase of risk  
5 characterization, that is, the risk description phase, which I  
6 will now discuss.

7           The second component of risk characterization  
8 is risk description where RQ values are calculated in the risk  
9 estimation phases are considered in conjunction with other lines  
10 of evidence which may either support or refute these estimates,  
11 based on assessment endpoints. In this part of the risk  
12 characterization, the relevance of other lines of evidence are  
13 considered.

14           The risk description builds on the RQ values  
15 calculated during the risk estimation phase of the risk  
16 characterization process. The risk description includes a  
17 discussion of possible refinements and exposure values --  
18 discussed by both Chris Garber and Reuben Baris -- that lead to  
19 refined RQ values, using toxicity estimates for individual and  
20 adult bees.

21           The quantitative information is then  
22 considered in the context of available studies that were  
23 discussed by Joe DeCant of the whole colony that may have been  
24 conducted under relatively controlled conditions, that is,  
25 semi-field studies, or under conditions that are intended to be  
26 more reflective of actual use conditions, that is, the full



1 field Tier III studies.

2 While adverse effects may be measured on  
3 individual bees and these effects quantified through risk  
4 quotients, the higher tier refinements are intended to answer  
5 whether adverse effects are observed at the whole colony level.  
6 The intent is for the risk assessor to determine whether or not  
7 there is a concordance of information to indicate whether there  
8 is a potential for adverse effects that could limit the extent  
9 to which the protection goals may be met.

10 Thus, while RQ values may indicate that  
11 individual bees may be at risk from acute exposures to a  
12 pesticide, semi-field studies may indicate that no adverse  
13 effects occur at the whole colony level when the pesticide is  
14 applied to pollinator attractive crop at the maximum proposed  
15 application rate.

16 Conversely, risk concerns, based on individual  
17 bees, may be supported by semi-field or full field studies,  
18 showing colony level effects. Other scientifically relevant  
19 information would be considered as well.

20 The risk description integrates multiple lines  
21 of evidence in the context of assessment endpoints discussed by  
22 Tom Moriarty in the opening presentation. As such, the risk  
23 description is not intended to be simply a comparison of risk  
24 quotients to the levels of concern, rather, the quantitative  
25 estimates of risk, that is, the RQ values are integrated with  
26 other lines of evidence that may be available through higher



1 tier guideline toxicity testing, non-guideline studies reported  
2 in open literature, and incident data.

3 Other lines of evidence, such as incident  
4 data, available through the Office of Pesticides Program  
5 Incident Data System, the IDS, and the Ecological Incident  
6 Information System, the EIIS, provide a means for understanding  
7 the effects of chemicals under actual use conditions.

8 As these additional lines of evidence are  
9 considered, the risk hypothesis and the conceptual model are  
10 refined. As discussed in the preceding presentations, the  
11 intent of the tiered risk assessment process is to provide  
12 increasingly realistic understanding of potential risk that  
13 reflects how the chemical will actually be used.

14 For new chemicals, available data may be  
15 confined to guideline laboratory and field testing; however, for  
16 older chemicals, a wider array of data may be available and are  
17 considered, provided those data meet the standard established by  
18 EPA for inclusion in risk assessments. EPA has developed  
19 guidance for evaluating multiple lines of evidence, and an  
20 example of such guidance is discussed in the White Paper where  
21 elements of the endocrine disrupting screening program, weight  
22 of evidence document are articulated.

23 Also, the Agency's risk characterization  
24 handbook developed by the EPA Office of Research and  
25 Development, also provides guidance on the factors that should  
26 be considered in risk characterization and which are discussed



1 in the following slides.

2 In considering the additional lines of  
3 evidence that may be available, the evaluation considers 1) the  
4 adequacy and quality of the data; 2) the degree and type of  
5 variability and uncertainty; and 3) the relationship of the  
6 evidence to risk assessment endpoints.

7 With respect to the adequacy and the quality  
8 of the data, risk assessors consider whether the data quality  
9 objectives were followed. OPP has published formal guidance on  
10 the conduct and evaluation of guideline toxicity studies and has  
11 developed guidance for staff in evaluating the utility of open  
12 literature studies that may not adhere to testing guidelines.

13 Just as the White Paper reflects and is  
14 consistent with the Agency's commitment to transparency, each  
15 risk description articulates what assumptions are made, where  
16 there are data gaps and what type of limitations may be present  
17 when non-guideline or supplemental data are used. As such, the  
18 experimental designs have been considered when evaluating the  
19 utility of the data for characterizing risk.

20 As was discussed in the presentation by  
21 Christina Wendel, the Agency is clear as to what is considered  
22 in the risk assessment process and what is not considered and  
23 why. Also, when evaluating lines of evidence, degree and type  
24 of uncertainty have to be articulated.

25 With respect to characterizing the degree and  
26 type of uncertainty and variability, variability refers to the



1 differences in characteristics of individuals or exposure,  
2 reflecting inherent variety in test organisms or exposure  
3 scenarios that can be quantified.

4 With respect to characterizing uncertainty,  
5 the risk characterization should include a discussion of what is  
6 not known, that is, of the uncertainties that may be associated  
7 with the available data. Also in evaluating lines of evidence,  
8 the relationship of the evidence to risk assessment has to be  
9 articulated.

10 Relationship of available evidence to risk  
11 assessment can draw from previous assessments on the pesticide  
12 or similarly structured pesticides and/or risk assessments  
13 conducted by other organizations, such as Pest Management  
14 Regulatory Authority -- or Agency, the Australian APVMA, and the  
15 European Food Safety Authority. Plus, the risk characterization  
16 must reflect current agency policy regarding the use and  
17 interpretation of data.

18 As discussed earlier by Tom Moriarty, the risk  
19 assessment framework discussed in the White Paper is intended to  
20 apply to honey bees; however, given that laboratory studies are  
21 based on individual bees and that higher tier studies, discussed  
22 by Joe DeCant examine both individual bees and colony level  
23 effects, the assessment may provide insight on potential  
24 exposure to and effects of the pesticide to solitary and social  
25 non-Apis bees as well.

26 In addition to measurement endpoints that are



1 directly related to assessment endpoints of a impaired growth,  
2 survival and reproduction, the risk description would also  
3 provide a discussion of sublethal effects that may have been  
4 measured in the various studies that have been evaluated, and  
5 the uncertainties associated with how these measurement  
6 endpoints may be related to the assessment endpoints.

7 Finally, the risk description should, to the  
8 extent possible, discuss the potential adversity of risks in  
9 terms of the nature and intensity of the effects, where there is  
10 evidence to suggest temporal or spacial trends, and whether the  
11 data indicate that recovery may be possible.

12 In the following presentation by Reuben Baris,  
13 the relative conservatism of the proposed risk assessment  
14 process will be briefly discussed. Also, as multiple lines of  
15 evidence are intended to be covered in the risk description, the  
16 white paper discusses the potential utility of colony level  
17 models to estimate potential effects of pesticides. Therefore,  
18 Keith Sappington will provide a presentation on how these models  
19 can inform the risk assessment process. Any questions?

20 **DR. DANIEL SCHLENK:** Wow. Thanks, Dr.  
21 Steeger. Any questions for Dr. Steeger? Dr. Pistorius?

22 **MR. JENS PISTORIUS:** I have one. Has there  
23 been an exercise done with a new proposed risk assessment  
24 process? How many substances would fail or pass compared to the  
25 old risk assessment? Is it many more or few more or all?

26 **DR. THOMAS STEEGER:** This is Tom Steeger.



1 That is a most excellent question and you're going to get an  
2 answer to it in the next presentation.

3 **DR. DANIEL SCHLENK:** Any other questions?

4 Okay. We're running a bit behind. I want to take a poll of the  
5 panel here. Who would rather take about a five minute break or  
6 keep going? We have two more presentations left to go. So, in  
7 favor of keep going, hands; and five minute break? Okay. Looks  
8 like we're going to keep going. Okay. With that, Reuben Baris,  
9 I guess you're up next.

10 **MR. REUBEN BARIS:** Thank you very much. My  
11 name is Reuben Baris. The information presented in this  
12 presentation was not included in the White Paper, but is  
13 intended to provide contextual information about the efficiency  
14 of the Tier I screen as presented in earlier presentations. As  
15 presented earlier, the purpose of the Tier I screen is to  
16 efficiently identify those pesticides that do not pose a risk to  
17 honey bees, so that researches can be focused on pesticides that  
18 potentially have risks.

19 Available acute oral and contact toxicity for  
20 adult worker bees from unpublished registrant studies were  
21 compiled for this analysis. This is a partial examination and  
22 is based only on available toxicity data for adult worker bees.

23  
24 Individual chemical risk quotients were  
25 calculated and compared to the proposed level of concern that  
26 was presented just now by Dr. Steeger. This effort was



1 completed to provide some context on the efficiency of the  
2 initial exposure screen using the most conservative assumptions  
3 of exposure. The Tier I screen for foliar applications were run  
4 for each chemical using the maximum foliar application permitted  
5 on the label. The Briggs model was not included in this  
6 analysis because there is limited oral toxicity information for  
7 adult worker bees for chemicals where soil applications are  
8 permitted on the label. Therefore, this analysis applies to  
9 foliar applications only.

10 A subset of adult worker acute contact  
11 toxicity data that cover a breadth of chemical categories,  
12 chemical groups and modes of action were used in this analysis.  
13 Shown here, the number of chemicals in each category is similar,  
14 and a few examples of the types of chemical groups are shown in  
15 the parentheses.

16 An attempt was made within each category to  
17 use chemicals from different groups and modes of action in order  
18 to remove potential bias towards a specific chemical activity.  
19 All available data from unpublished registrants submitted acute  
20 oral toxicity studies for adult worker bees were used in the  
21 acute oral analysis. Many of the chemical groups and modes of  
22 action represented in the acute contact analysis were also  
23 represented in this acute oral analysis.

24 This table and the following slide represent a  
25 snapshot of the Tier I screen based on available acute contact  
26 toxicity data. As was presented by Kris Garber this afternoon,



1 the Koch and Weisser approach with honey bee specific data is  
2 the agency's proposed approach for the Tier I contact exposure  
3 screen.

4 Shown here, fewer insecticides make it through  
5 the screen and would likely require additional refinement.  
6 However, herbicides and fungicides generally pass the screen  
7 with approximately 32 and 4 percent respectively requiring  
8 additional evaluation or refinement.

9 Fewer insecticides pass the initial Tier I  
10 screen compared to herbicides and fungicides. This is expected  
11 because insecticides are more likely to directly affect insects  
12 such as honey bees. It is clear from the table shown there that  
13 while greater than 85 percent of the insecticides evaluated did  
14 not pass the screen, the evaluation was based on acute toxicity  
15 studies with young adult bees.

16 Larval toxicity testing may result in RQ  
17 values that exceed the level of concern of 0.4. However, even  
18 for some insecticides, it is possible that bees are not  
19 particularly sensitive, and these chemical will likely have RQ  
20 values that are below the LOC, just as not all plants are  
21 sensitive to herbicides.

22 Since fungicides and herbicides tend to target  
23 fungi or plants, it is less likely that they will directly  
24 affect insects. This analysis was also completed with the T-REX  
25 contact approach, and similar results were observed.

26 Again, as was expected in this acute oral



1 analysis for adult worker bees and shown previously on the  
2 previous slide by the contact exposure analysis, fewer  
3 insecticides pass the initial Tier I screen compared to  
4 herbicides and fungicides. There are a limited number of  
5 fungicides that are included in this analysis because of limited  
6 oral toxicity data that are available for adult worker bees.  
7 But again, as shown by these preliminary results, the Tier I  
8 screen is functioning well as a screen by identifying the  
9 chemicals that need additional refinement and screening out the  
10 majority of non-insecticides that are not likely to be a risk  
11 concern.

12 This analysis was an exercise to illustrate  
13 the decision point of Box 5(a) and (b) of the proposed decision  
14 tree. That is, does the contact or oral RQ exceed the level of  
15 concern? It provides context for the efficiency of the Tier I  
16 screen. The analysis included a variety of chemical groups and  
17 modes of action for contact and oral toxicity, identifying those  
18 chemicals that require additional refinements. These  
19 refinements, Box 6 of the decision tree would include working  
20 with risk managers to refine the Tier I exposure assessment.

21 Therefore, the conclusion is that the Tier I  
22 approach is functioning as expected as an initial screen. It is  
23 important to keep in mind that these preliminary results are  
24 intended to give a very rough example that the screen is not  
25 serving as a wall for all classes of pesticides, but rather, it  
26 is identifying those chemicals that are most likely to need



1 additional review. In doing so, only model-estimated exposure  
2 values have been evaluated.

3 It is possible that measured exposure values  
4 could result in a reduction in risk estimates such that further  
5 refinements -- that is higher tiered testing -- may not be  
6 required. However, as mentioned by Dr. Steeger in the previous  
7 presentation, multiple lines of evidence are used in determining  
8 the risk to honey bees.

9 The next presentation given by Mr. Sappington  
10 will discuss colony level models and how they can be used in the  
11 risk characterization process. Thank you for your time.

12 **DR. DANIEL SCHLENK:** Questions? Dr.  
13 Berenbaum?

14 **DR. MAY BERENBAUM:** While fungicides might  
15 themselves not be considered to present as greater risk, there  
16 is increasing evidence that in combination fungicides can  
17 exacerbate risks. Were any studies conducted on combinations or  
18 synergistic interactions?

19 **DR. THOMAS STEEGER:** This is Tom Steeger.  
20 That as Reuben pointed out towards the end of his presentation,  
21 the risk quotient isn't viewed in isolation and other lines of  
22 evidence would be considered. And you're absolutely right,  
23 there is open literature pointing to potential interactive  
24 effects and we have required studies to address those  
25 uncertainties, even for chemicals that had initially RQ values  
26 that would have been below the LOC.



1                   **DR. DANIEL SCHLENK:** Okay. Any other  
2 questions? Yeah, Dr. Hunt?

3                   **DR. GREG HUNT:** Well, along those same lines,  
4 some formulations come as a package, such as seed treatments.  
5 Is there any consideration that that might be required of  
6 registrants to - I know you can't test every possible  
7 interaction, but there are some common formulations that could  
8 be tested.

9                   **DR. THOMAS STEEGER:** There is a terminology  
10 difference that we deal with. When we talk about formulations,  
11 oftentimes what the agency is meaning is chemicals that are  
12 co-formulated into a single product as opposed to what you might  
13 see happen out at the field level where are tank mixes, and  
14 these aren't really formulations. These are brews, for lack of  
15 a better term, that are used.

16                   Where we have an indication that a particular  
17 formulation could potentially be more toxic to a taxa, whether  
18 it be honey bee or any taxa that we're evaluating, we may call  
19 in - again it's a risk management call - data to address that  
20 uncertainty.

21                   **DR. DANIEL SCHLENK:** Okay. No further  
22 questions? Our last presentation of the day, no pressure there  
23 Keith.

24                   **MR. KEITH SAPPINGTON:** Thanks for bringing up  
25 brew. This will make me go faster.

26                   Good afternoon. Again, my name is Keith



1 Sappington with the Environmental Fate and Effects Division. I  
2 will be changing gears a little bit here in the context of our  
3 previous discussions and talking about the potential use of  
4 mathematical models in the context of pesticide risk assessment  
5 for honey bees.

6 For convenience, we've termed these as colony  
7 level models, which mean that the models focus on describing the  
8 dynamics of a single colony, not a population of multiple  
9 colonies. This information is found in section 5.4 of the White  
10 Paper. I would also like to acknowledge the work of Kris Garber  
11 who helped greatly in the review, providing additional support  
12 for this topic in the White Paper.

13 So, first, I will briefly describe some of the  
14 reasons why we may be interested in colony-level models in the  
15 context of pesticide risk assessment with honey bees and then I  
16 will discuss their current state of the science. Finally, I  
17 will conclude with a brief description of selected models that  
18 we identified in the literature.

19 There are a number of reasons why conceptually  
20 we would be interested in these models, and I'm sure they are  
21 obvious to the panel at this point. First of all, these models  
22 enable the dynamics of a honey bee population to be measured.  
23 These dynamics is shaped numerous interdependent processes,  
24 which can be partially informed and managed through biological  
25 feedback loops.

26 For example, excessive loss of foragers may be



1 partially compensated for by an earlier recruitment of hive bees  
2 into the forager workforce. Such a change may compensate for  
3 the loss of foragers in the short term, but may in the longer  
4 term lead to adverse effects on the colony through a reduction  
5 in brood rearing efficiency as well as a reduction in worker  
6 longevity.

7 While empirical studies may capture the net  
8 effect of these interdependent processes on overall colony  
9 strength, a properly constructed colony-level model could  
10 provide a means for understanding these relationships, and  
11 therefore enable one to extrapolate observed effects over  
12 perhaps longer period of time and possibly under different  
13 environmental conditions.

14 Second, colony level models could help in  
15 interpreting sublethal effects, which we have discussed on  
16 repeated occasions in earlier presentations. And in particular,  
17 for linking sublethal effects quantitatively on individual bees  
18 to effects at the whole colony level.

19 Third, colony level models could also help in  
20 a design of empirical studies such as the Tier II and Tier III  
21 field studies described by Joe DeCant. For example, they could  
22 be used in addressing study design elements such as when to  
23 conduct a study, at what spacial scale, and even help with  
24 deciding on the degree of replication necessary. They also can  
25 help in identifying the duration that may be needed for a  
26 particular semi-field or field study.



1 Fourth, such models could also assist in  
2 interpreting the results from the Tier II and Tier III studies,  
3 such as accounting for the seasonal effects on measurement  
4 endpoints or other factors like the impact of the weather on  
5 model results, or on observed results.

6 In the context of the proposed risk assessment  
7 scheme, colony-level models could enable one to interpret  
8 results of toxicity studies and integrate these results from  
9 different risk assessment tiers, all the way from Tier I at the  
10 individual level through Tier II and Tier III.

11 Finally, such models could potentially  
12 incorporate not only the effects of a single stressor, but those  
13 of multiple stressors, which may be either chemical or  
14 nonchemical in nature.

15 The overall state of the science of  
16 colony-level models for honey bees was recently reviewed and  
17 published as part of an executive summary of the SETAC Pellston  
18 workshop. We've included the conclusions of this review here  
19 because it was conducted specifically in the context of their  
20 potential application in pesticide regulatory risk assessment.

21 In this review, it was concluded that none of  
22 the available honey bee models seemed suitable for regulatory  
23 risk assessment for several reasons listed here. Collectively,  
24 these include a lack of explicit linkage of foraging bees to  
25 their surrounding landscape, insufficient testing of model  
26 results against empirical data, lack of a comprehensive



1 sensitivity analysis in order to understand the impact of model  
2 parameters and assumptions, exclusion of multiple stressor  
3 impacts on model results and insufficiency documentation of  
4 model assumptions and/or algorithms in some cases.

5 At this time, only the SETAC executive summary  
6 is available from the Pellston workshop. So, the detailed  
7 review specific to each model was not available to us.

8 Because the details were not available at the  
9 time we were drafting the White Paper, we decided to conduct a  
10 very brief review of only selected models that we identified in  
11 the literature. As we point out in the White Paper, this review  
12 is not intended to be comprehensive or exhaustive. Instead, it  
13 reflects a sampling of more recent models that had been  
14 published for honey bees as a way for us to explore their  
15 potential utility in pesticide risk assessment.

16 In this and the subsequent table, we summarize  
17 some of those attributes of the models we reviewed. The models  
18 are listed here from top to bottom, alphabetically by the lead  
19 author of the publication in which they appeared, and various  
20 attributes are listed at the top of this table. We note that  
21 inclusion of a model in this list does not constitute our  
22 endorsement nor does exclusion imply lack of endorsement.

23 So, in looking at the first two columns, one  
24 can see that all but one of the models we considered address  
25 effects at the colony level with output including overall colony  
26 strength in composition. The model published by Makela et al.



1 in 1993 is a true population model for honey bee in that it  
2 considers multiple honey bee colonies since it was designed to  
3 predict the impact and distribution of Africanized bees on the  
4 European honey bee.

5 In the third column of this table, I've  
6 included some key model input parameters and model processes.  
7 I've also provided some of the unique features of each model in  
8 bold. For example, the model by Becher et al. in 2010 shown in  
9 the first row focuses on the distribution of comb temperature  
10 and how this in turn affects queen egg laying rate, brood  
11 development and overall colony strength. These authors  
12 acknowledge that the their model is, in effect, a partial model  
13 and it's not intended to address overall colony dynamics.

14 Among other features, the BeePoP model  
15 published by DeGrandi-Hoffman shown in the second row includes  
16 an explicit representation of weather patterns in addition to  
17 modeling queen fertility as a function of queen age.

18 The model by Khory et al. shown here in the  
19 third row was specifically designed to evaluate age-related  
20 division of labor among hive bees that is related to the onset  
21 of foraging. This model was designed to investigate  
22 specifically how the early onset of foraging could impact the  
23 survival of foragers as well as brood rearing efficiency.

24 The inter-colony model, I just mentioned by  
25 Makela et al. in the fourth row contains some unique attributes  
26 such as bee swarming, inter-colony migration of bees and even



1 changes in genotype.

2 Perhaps the most comprehensive colony model in  
3 this collection in this table is the HoPoMo model published by  
4 Schmickl and Crailsheim in 2007. This model listed in the fifth  
5 row includes, among many other processes, foraging activity,  
6 food availability and even larval cannibalism and their impact  
7 on colony strength and composition.

8 Lastly, the model in the last row by Thompson  
9 published in 2005 focuses on pesticide impacts on honey bee  
10 processes, including the queen egg laying rate, the onset of  
11 foraging activity, brood rearing and worker longevity.

12 Other features of these models that we  
13 summarize include whether or not it is able to accommodate  
14 spatially explicit information, whether a formal sensitivity  
15 analysis was performed, the extent to which it has been verified  
16 using empirical data and its application to pesticide risk  
17 assessment.

18 Several models we looked at were able to  
19 include some spatially explicit or geographic based information,  
20 although the BeePoP and HoPoMo models, this inclusion is limited  
21 to climate data, which can be made region or site-specific.  
22 Most models did not include a formal sensitivity analysis except  
23 for the BeePoP model, although some limited analysis of selected  
24 model input parameters was conducted in some other studies.

25 In terms of verifying model projections with  
26 empirical data, only some of the models included a comparison of



1 model results or model components to observe data. For example,  
2 Becher et al. compared their predicted timing of peak hive  
3 strength and minimum number of bees necessary for successful  
4 overwintering of hives to observe values.

5 Khoury compared a predicted onset of foraging  
6 and the forager age to literature values. For the HoPoMo model,  
7 Schmickl and Crailsheim compared several model subcomponents as  
8 well as the entire model output. That is a number of bees to  
9 observed data.

10 Lastly, only two of the models that we  
11 identified in this list included an explicit application to  
12 pesticide risk assessment.

13 In this slide, we thought it would be useful  
14 to articulate some of the main attributes that we believe are  
15 desirable in order to apply a colony-level model in pesticide  
16 risk assessment. As indicated in the first bullet, it's is  
17 clearly important to account for the major processes that govern  
18 the dynamics of honey bee colonies, initially at the colony  
19 level and eventually at the population of inter-colony level.

20 Second, the model should quantitatively link  
21 the various measurement endpoints from pesticide toxicity  
22 studies to the proposed assessment endpoints. For example,  
23 colony strength and survival, production of hive products.

24 As indicated in the third bullet, such models  
25 should be able to be readily parameterized using existing  
26 biological and pesticide-specific exposure and effects data.



1 Another desirable feature shown in the fourth  
2 bullet is the ability to tailor model results to a specific  
3 region of the country in order to account for differences in  
4 pesticide use patterns, crops, climate and even landscape  
5 features.

6 Lastly, but no less important, the model must  
7 be scientifically defensible, highly transparent, well  
8 documented and publically available in keeping with the Agency's  
9 efforts to promote transparency with clear, concise and  
10 reasonable risk characterizations and tools used therein.

11 We note in the footnote that EPA has published  
12 guidelines for the development of environmental models in  
13 general that are intended for use in regulatory activities. In  
14 addition, the European commission is developing several model  
15 guidelines specifically for mechanistically-based ecological  
16 effects models.

17 Among other factors, a colony-level model  
18 should include a formal sensitivity analysis and uncertainty  
19 analysis in addition to verification of the model results to  
20 observe empirical data.

21 So to conclude, the SETAC Pellston workshop  
22 participants indicate the currently colony level simulation  
23 models are not ready for regulatory application. This  
24 conclusion seems consistent with our cursory review of a limited  
25 subset of models. However, once these models are fully  
26 developed and evaluated, we think the colony-level models could



1 provide an important line of evidence in the risk  
2 characterization of pesticide effects on honey bees, and  
3 potentially help to identify adverse outcome pathways across  
4 multiple levels of biological organization.

5 As we pointed out, they could be extremely  
6 valuable in the problem formulation phase in an assessment in  
7 the design and interpretation of field studies and in  
8 integrating pesticide effects and exposure data generated at  
9 multiple risk assessment tiers and at various levels of  
10 biological organization.

11 Therefore as indicated in the final charge  
12 question number 14, we are interested in SAP feedback on the  
13 concept of using colony-level models in pesticide ecological  
14 risk assessment. In other words, is this concept worth pursuing  
15 by the pesticide regulatory agencies represented?

16 In addition, we would like SAP feedback on the  
17 current state of the science of honey bee models in relation to  
18 their potential use and application in pesticide risk  
19 assessment.

20 And finally, we would appreciate your input on  
21 the most important model design elements that we should consider  
22 as part of any review of a colony level model for application in  
23 pesticide risk assessment. Thank you very much.

24 **DR. DANIEL SCHLENK:** Any questions? Dr.  
25 Fefferman?

26 **DR. NINA FEFFERMAN:** Just as a very high-level



1 cursory question in shifting from the White Paper discussion to  
2 your presentation today, there was a shift from a discussion of  
3 general models for colony-level use to simulation models. I was  
4 wondering if that was purposeful and if you were interested in  
5 analytic mathematical models and formulations as well as  
6 simulation-based methods?

7 **MR. KEITH SAPPINGTON:** Could I ask you to  
8 clarify general models as compared to mathematical models?

9 **DR. NINA FEFERMAN:** Sorry, so in my mind sort  
10 of mathematical analytic models live on one side and simulation  
11 models live on another. So, simulation models are ones where  
12 there are algorithms for action, and from those you get  
13 numerical solutions, whereas mathematical models are ones where  
14 by understanding the interactions of systems of equations, you  
15 do things like generate threshold behavior or equilibrium  
16 states. Things that don't involve, for example, running Monte  
17 Carlo analyses in the same way as outcome measurements.

18 So, simulation models produce data. Analytic  
19 models produce sort of dynamic insight, and I was wondering if  
20 you are specifically restricting yourself to simply the  
21 simulation methods or if you're also interested in methods that  
22 provide close form equation type understanding.

23 **MR. KEITH SAPPINGTON:** Keith Sappington.  
24 There was not an intentional restriction to the simulation  
25 models, although they were attractive because of the analytical  
26 components and the type of measuring endpoints included and the



1 outcome aligning very closely to our assessment endpoints. With  
2 that said, if the analytical versions of the models could  
3 provide us insights, for example, the thresholds that need to be  
4 maintained for long-term hive success, we would welcome input on  
5 that.

6 **DR. NINA FEFFERMAN:** Thank you. I'm sure I'll  
7 talk about this more with you when we get to the specific  
8 modeling section of the discussion, but thanks. That's great.

9 **DR. DANIEL SCHLENK:** Dr. Schwab?

10 **DR. PAUL SCHWAB:** Given the fact that the  
11 number of times we have talked about chronic effects, one of the  
12 models, I think it was the last one in your table, was talking  
13 about the impact of pesticides in sort of a chronic fashion  
14 rather than in terms of mortality, on laying of eggs and so on.  
15 What was the basis of that model if there is really a lack of  
16 data to put that together?

17 **MR. KEITH SAPPINGTON:** Are you referring to  
18 the Thompson model?

19 **DR. PAUL SCHWAB:** Yes I am.

20 **MR. KEITH SAPPINGTON:** That was built on - in  
21 that paper, the authors actually coupled semi-field experiments  
22 on colonies and used that information as input into their -- it  
23 was actually a colony model or, excuse me, a model developed for  
24 Varroa mites, actually predicting Varroa mite infestations.  
25 They adapted that model, which was published earlier, and then  
26 included information from their colony-level tests as well as -



1 I think, I'll have to check - individual level information from  
2 various pesticides, and then looked at the results from that,  
3 both in terms of the empirical studies as well as the  
4 simulation.

5 **DR. PAUL SCHWAB:** So, the effect of the  
6 pesticide was a negative impact on the bees from the pesticides  
7 themselves, or somehow the pesticides interacting to take care  
8 of the mites that they were originally looking at?

9 **MR. KEITH SAPPINGTON:** Keith Sappington. They  
10 did not include mites in this particular simulation. The model  
11 itself was derived from a model built to investigate mite  
12 infestations, but they adapted that and then coupled measured  
13 data on colony level effects, and basically tried to compare the  
14 two.

15 **DR. DANIEL SCHLENK:** Okay. Dr. McManaman?

16 **DR. JAMES MCMANAMAN:** Yeah, I wonder if we  
17 could bring up Mr. Moriarity's presentation. I have a question  
18 that's related to models, but it's more related to population of  
19 bees. Is that possible?

20 **DR. DANIEL SCHLENK:** They are working on it.  
21 Is there another question that we can do while they're bringing  
22 that up? Does somebody have something else that we can do? Oh,  
23 he's got it.

24 **DR. JAMES MCMANAMAN:** It's number four. So, I  
25 was struck by this, and I didn't know exactly - it didn't seem  
26 like it was an appropriate time to ask during the presentation.



1  
2 This is kind of an unusual curve in that it  
3 looks like there is a steady decline from about 1950 to about  
4 1970 in the pollinator population. Then, there's this plateau,  
5 kind of up and down plateau for about 20 years, and then when  
6 the mites hit, there is a sharp decline. Then, from you know,  
7 1995 to about 2006, it looks like there that there is another  
8 plateau here.

9 So, this looks like there is a multivariable  
10 interaction going on with the pollinator population in general.  
11 Has there been any kind of analysis to try to attempt to look at  
12 potential -- kind of like an epidemiological analysis -- to look  
13 at what was going on? Because in order to understand what could  
14 contribute to pollinator decline would really be nice to know  
15 what environmental changes have happened, what other kinds of  
16 factors could have gone into the declines that we have seen  
17 historically.

18 And part of this question is that this  
19 plateau, the last plateau, it looks like there is a bit of a  
20 decline at about 2005/2006 - do you have more recent data on  
21 that the know does that continue to go down or is that just kind  
22 of a blip and it really steadies off?

23 **DR. THOMAS STEEGER:** This is Tom Steeger. A  
24 couple of comments about this graph. I think that you need to  
25 be careful in terms of interpreting it as it speaks to  
26 pollinator declines per se. It's certainly informative about



1 the issue of pollinator declines. This is a graph of the number  
2 of commercial colonies that are used in honey production in the  
3 United States.

4 What it is probably more reflective of is a  
5 changing demographic; that after the war there wasn't the need  
6 for people to produce sweetener because sugar was in short  
7 supply. And so, more and more people got out of the business of  
8 honey production and the other part of this, it's hard to say  
9 how much of an impact it had, was on the way that NASS actually  
10 collected data, and their exclusion of sort of back yard  
11 beekeepers that are producing honey to more commercial honey  
12 production is an artifact that is part of this graph. So, it  
13 doesn't really present what might be represented -- the  
14 contribution from actual pollinator declines. So you have to be  
15 careful with how you interpret this graph.

16 As was pointed out in the presentation, it's  
17 not - this graph goes out to 2006. The peak was about roughly 6  
18 million colonies. It was actually 5.5 million in the late  
19 1940s. The most current estimate, the 2011 estimate, places the  
20 number of managed honey bee colonies for honey production at  
21 2.49 million. So, you still see this decline taking place. As  
22 was discussed in Tom Moriarity's presentation, these losses that  
23 are being reported by beekeepers of 30 to 34 percent and the  
24 high 20s this year because of a mild winter, these are real  
25 losses. It's not just changing demographics.

26 There is something happening that is really



1 difficult to understand in terms of what combination of factors  
2 are actually causing it. But getting to your question about  
3 epidemiological approaches, it's interesting you should bring  
4 that up because Rosalind James certainly has proposed that to  
5 potentially pull in super-computing capabilities to get at that  
6 issue. It's certainly an issue that USDA has been tasked with  
7 over the past five years, congressionally mandated to determine  
8 causes, and the week after next, there is actually a conference  
9 to look at causal analysis on what is causing bee decline.

10 So, there is a lot of interest and energy  
11 being put at addressing this. It certainly was an issue that  
12 was flagged by NAS in their publication on pollinator declines  
13 in North America, but relative to this graph in terms of  
14 interpreting plateaus and the peaks, it can be difficult to  
15 interpret. It looks like Dr. Berenbaum has something to add.

16 **DR. MAY BERENBAUM:** Yeah, this was a subject  
17 of discussion of the NRC, a study on the status of pollinators  
18 in North America. Just to amplify what Dr. Steeger said, this  
19 is only, it's not pollinator decline. It's honey producer  
20 decline. These data don't reflect the changing nature of the  
21 apiculture industry with a greater emphasis on provision of  
22 pollinator services. It's just honey production up through  
23 2006. That steep drop in the mid 1980s also, although it's easy  
24 to attribute that drop to the introduction of mites, it  
25 coincides with the change in methodology. So, there is a  
26 confounding factor there.



1           As you said, the sideliners, hobbyists, anyone  
2     with five or fewer hives is not included. So, it's not suitable  
3     for epidemiological analysis. It's really reflective more of  
4     the industry than it is of the pollinator workforce of the  
5     United States. You know, it's hard to over interpret because it  
6     was not initially set up for that purpose. It was more for  
7     honey production.

8           **DR. JAMES MCMANAMAN:** This is Jim McManaman  
9     again. So, what is the state and in terms of assessing the role  
10    of pesticides and their contributions to decline, whether this  
11    is through honey production, but in terms of overall pollinator  
12    decline, it would be helpful to really have an accurate idea of  
13    what the real nature of the decline has been because it sounds  
14    like this is maybe an artificial - it may not be reflecting  
15    reality. So that's the basis of my question. Is it still going  
16    down or has it plateaued off?

17          **DR. MAY BERENBAUM:** Maybe Jeff, Dr. Pettis  
18    could address the Apiary Inspector of America Effort to obtain  
19    better data.

20          **DR. DANIEL SCHLENK:** Go ahead, Jeff. Thanks.

21          **DR. JEFF PETTIS:** Very briefly, Jeff Pettis,  
22    USDA. As May pointed out, this is just managed honey bee, and  
23    actually there has been an analysis of the slope before that  
24    blip and afterwards that it's the same - it's mostly driven by  
25    economics, honey prices in fact.

26          As far as getting at the loss rate of let's



1 say honey bee colonies in the U.S., NASS was encouraged to take  
2 over and do a very large study and they had no funding to do  
3 that. There has been an effort for six years now, but we've  
4 done a survey of losses in the U.S. and we asked a number of  
5 questions.

6 We're looking at ways to probe that data and  
7 try to get at, you know, Varroa mites versus pesticide exposure,  
8 et cetera. It's not the best dataset, but it's the best dataset  
9 we have. We are looking at various ways to use that dataset to  
10 try to get at some of these multifactorial issues, but a lot of  
11 it is based on questionnaires where we have, depending on the  
12 beekeeper to give us truthful answers.

13 So, we have some other better datasets, but  
14 only for specific colonies, two to three hundred colonies, not a  
15 huge dataset.

16 **DR. DANIEL SCHLENK:** Okay. Any questions for  
17 the Agency? Thanks. Any questions? Dr. McManaman?

18 **DR. JAMES MCMANAMAN:** This is Jim McManaman  
19 again. So, I'm going to push this point just a little bit more.

20 So, in terms of understanding the risk and developing risk  
21 assessment, it really would be good to have numbers about if  
22 there are any inaccuracies and what the actual decline is, it  
23 really would in terms of setting policy and going forward on  
24 this, it doesn't sound like we really have the numbers in hand  
25 to really assess the real risk to the pollinators. Maybe I'm  
26 over stating that.



1                   **DR. THOMAS STEEGER:** This is Tom Steeger. I  
2 think that if there's any uncertainty in the room that  
3 pollinators, particularly the honey bees, where there is a  
4 question as to whether they are experiencing significant  
5 declines, as someone who is responsible for monitoring the  
6 incident database, I have received calls and talked to  
7 individual beekeepers that have lost within a period of two  
8 weeks 21,000 colonies. As a biological event, if you figure  
9 that there are 50,000 bees per colony, the loss of 21,000  
10 colonies in that short of a period of time is a staggering  
11 event.

12                   We are unclear as the role that pesticides are  
13 playing in pollinator declines. There have been multiple  
14 factors as was discussed in Tom Moriarity's presentation that  
15 USDA has identified as contributing to pollinator declines. Do  
16 we have a firm understanding of what declines are actually out  
17 there? I don't think so, because it's difficult to get a handle  
18 on this data, but I think that as an agency that is responsible  
19 for regulating pesticides, we have a responsibility to document  
20 to the best of our extent, the extent that pesticides are  
21 playing a role and to mitigate those effects to the extent  
22 possible.

23                   But it has been my experience based on  
24 information that I get through incident reports and through my  
25 interactions with the beekeeping community in the United States  
26 that these losses are significant, and people who have been in



1 the business of beekeeping who have an understanding of how to  
2 keep bees are experiencing staggering losses. This is not a  
3 contrivance. It is a real problem. The ability of the country  
4 to meet pollination service requirements is becoming a question,  
5 given the increased need for pollination services for  
6 pollinator-dependent crops.

7 **DR. JAMES MCMANAMAN:** This is Jim McManaman  
8 again. I certainly didn't want to imply that there was a  
9 contrivance, but you know, as a matter of it's just difficult to  
10 assess what the contributions of pesticides might be to the  
11 loss. There may be multiple factors leading to it. So, without  
12 hard numbers, it's just really, you know, we might be shooting  
13 at the wrong target is all I'm saying.

14 **DR. DANIEL SCHLENK:** Yeah, Dr. Fefferman?

15 **DR. NINA FEFFERMAN:** Something occurred to me  
16 very briefly. For the transparency and clarity aspects of the  
17 demands from the modeling side, transparent and clear to whom?  
18 To practitioners? To biologists? To beekeepers? To modelers?  
19 Where does that benchmark get set?

20 **DR. THOMAS STEEGER:** Presumably to the risk  
21 manager as the primary client of our risk assessment, but it's  
22 to the stakeholders as well who are affected by our decisions.

23 **DR. NINA FEFFERMAN:** So does that mean that  
24 the methodology by which it was arrived must also be clear  
25 because I think that might eliminate almost all modeling from  
26 being sufficiently transparent.



1                   **DR. THOMAS STEEGER:** As Keith Sappington  
2 pointed out, the Agency has specific guidance on the development  
3 of models and the transparency has to do with the coding that  
4 goes into the algorithms that are used to support those models,  
5 and by the testing that goes on subsequent to determine how well  
6 those model estimates reflect actual exposure or effects that  
7 occur at the colony level.

8                   **MR. KEITH SAPPINGTON:** Keith Sappington. I  
9 would add in addition to just having the equations in the Paper,  
10 having the basis for parameters that are used for those  
11 equations, and then again, having that information. So, then it  
12 could be evaluated by anybody in terms of a scientific  
13 defensibility.

14                   **DR. NINA FEFERMAN:** So, thank you. I mean I  
15 guess part of why I'm asking is that again, moving towards the  
16 more analytic side of models. There are occasionally very  
17 valuable insights that can be provided in the absence of data or  
18 validation in the sense of ground truthing outcomes, as long as  
19 the process itself is understood to be reflective of a realistic  
20 process. So, in some sense, there are no parameters that come  
21 in, there are simply functional relationships. And in that  
22 sense, then the equations can still provide quite a bit of  
23 insight into things like the existence of a meaningful threshold  
24 within, for example, this thing can't go negative, and therefore  
25 there exists no satisfiable threshold, because otherwise we've  
26 created bees out of thin air kind of validity.



1                   On that level, sometimes there can be English  
2 descriptions, but there isn't so much to grab on to. So, I'm  
3 wondering are there guidelines that apply equally well to models  
4 like that? Are they necessarily then off the table? What  
5 happens?

6                   **DR. STEPHEN BRADBURY:** Maybe there is another  
7 document we could try to get into the record, which may get a  
8 bit at what you're asking. There is an OECD document, and it's  
9 for quantitative structure activity relationships, which is a  
10 different specific topic, but it talks about model - it calls it  
11 model validation; but it is more around what kind of thing  
12 should you kind of describe about your model.

13                   It doesn't come at that a model is right or  
14 wrong, it's about describing what the model does and various  
15 attributes of the model and explaining it in such a way that a  
16 user of the model can be informed as to whether or not it's  
17 appropriate to use a given model for the given purpose that  
18 somebody maybe - or question somebody may be trying to answer.

19                   So, I don't know if it will match exactly what  
20 you're getting at, but I think it might provide some insights  
21 into where we're coming from. It isn't the model is good, bad  
22 or indifferent, it's how do you try to describe attributes of  
23 the model so that you can understand how to use it or not use it  
24 or use it with care for a given question you may need to answer.

25  
26                   Getting back to your transparency and



1 understandability, certainly it's in the risk assessment  
2 framework that the risk assessor and the risk manager hopefully  
3 understand each other so they're not passing in the night and  
4 burning resources in time, misunderstanding each other; but also  
5 at the end of the day it's to ensure that the public understands  
6 what it is we're thinking and how we formulated our position.  
7 Clearly, sometimes these documents are huge. So, we have a  
8 range of audiences from scientific experts that know the very  
9 excruciating detail of a certain component of the risk  
10 assessment and somewhere in that risk assessment document, being  
11 able to explain it to that audience. But ultimately being able  
12 to go through risk communication and analysis and writing so  
13 that public can understand the rationale to the decision making.  
14 So it gets challenging, but that's what we're trying to strive  
15 for.

16 **DR. NINA FEFFERMAN:** Thank you, that sounds as  
17 though the extra document you were thinking of would get to  
18 exactly the sort of thing that I'm thinking, and it does also  
19 set a useful benchmark for me in considering. Thank you.

20 **DR. DANIEL SCHLENK:** Dr. Pettis?

21 **DR. JEFF PETTIS:** More of a comment, but it  
22 does tie into the modeling, and it gets back to your question a  
23 bit.

24 As you go from Tier I to Tier II or Tier III,  
25 you go from control studies with individual bees, which we can  
26 control. We can control the genetics. We can control



1 everything. Then you get an interpretation of some result of  
2 these individual bees. As soon as you take it Tier II or Tier  
3 III, where you have colony level, you bring in all this  
4 uncertainty. I think that's what we grapple with.

5 I mean, so maybe a modeling approach,  
6 especially at the colony level, it can be very useful and maybe  
7 applying a certain degree of loss or something attributable to  
8 one thing and then following that over time. But as soon as you  
9 go to the colony level, it's impossible to control all these  
10 other factors, and that's what we grapple with.

11 **DR. DANIEL SCHLENK:** That would be a great  
12 discussion for tomorrow and the next day. Looking for any other  
13 questions for the Agency while we got them here. Yes, Dr.  
14 Potter?

15 **DR. THOMAS POTTER:** This is Tom Potter here.  
16 We've been looking at question 14 because I was identified as an  
17 associate discussant on there. So, I have been trying to get my  
18 head around it. I think it's question 14 and it has to do with  
19 a colony-level modeling. I thought the answer was easy. Yes,  
20 it's a great thing. Boy, I wish we had the simulation model  
21 where we could assess these effects, but apparently it doesn't  
22 exist and they exist in various, what appear to be crude form.

23 So, ultimately my question comes around to you  
24 folks is you know, what kind of response are you looking for?  
25 Is the answer yes, models are a great idea? I think that one's  
26 easy. And the second part of it is who is going to develop



1 those models and support them? Obviously, that's a  
2 fundamentally important question with regard to any simulation  
3 model that we might use.

4 **MR. KEITH SAPPINGTON:** Keith Sappington. I  
5 think the first thing we wanted was the simple question, perhaps  
6 yes, was that is this concept worth dedicating some amount of  
7 resources to, because it is a rather large effort even to take  
8 an existing model and understand the transparency, understand  
9 all of the information that went into it, develop it and modify  
10 it and then bring it to a panel like this for final review.

11 So, before doing that and in the context of  
12 pollinator declines and multiple stressors, we wanted to just  
13 check in before considering this option further.

14 The second thing is getting sort of a check in  
15 on this state of the science. Do you agree that with the  
16 current state of the science, what we were able to do just  
17 briefly was to summarize a few of the models.

18 And then thirdly, sort of helping us evaluate  
19 or even perhaps dedicate efforts to developing some models,  
20 understanding what are the most important factors to include in  
21 a model?

22 The HoPoMo model has 66 differential equations  
23 within it, and all models are wrong and some models are useful.  
24 So, I'm not sure that we need a model with 66 differential  
25 equations. So, the advice that would be very helpful to us  
26 would be to say, well here's your top tier of things that you



1 must include in a model to have any chance of getting a  
2 realistic result, and then here are some other sort of secondary  
3 tiers that gee, if you have the data, this will be useful and it  
4 will be useful for this purpose.

5 **DR. DANIEL SCHLENK:** Dr. Berenbaum?

6 **DR. MAY BERENBAUM:** Not to prolong this, but  
7 just to follow up and reflect my profound ignorance of how  
8 Washington, DC, works. So, we are likely to identify data gaps,  
9 but isn't EPA primarily regulatory and whose responsibility is  
10 it then to fill those data gaps? Is it kind of who will help me  
11 bake the bread scenario. Yeah, how does our response to the  
12 charges figure into meeting the EPA's need for certain types of  
13 information?

14 **DR. DANIEL SCHLENK:** Dr. Bradbury, I think  
15 would answer that one, yeah?

16 **DR. STEPHEN BRADBURY:** I will just be careful  
17 on my editorial comment about Washington. So, it gets back to  
18 some of the earlier questions in terms of will it go quick. But  
19 the big picture, what's the cause of pollinator decline. That's  
20 an activity, USDA is leading it and multiple federal agencies  
21 are involved in academia. So, as we said at the beginning of  
22 today's discussion, we're focusing on the pesticides first in  
23 terms of under FIFRA we do take a look at effects of pesticides  
24 on nontarget organisms, pollinators are an important component  
25 in the ecosystems. They have benefits for agricultural  
26 production and the products of the hive, with everything else



1 that's going on, making sure we're advancing our capability of  
2 doing that.

3 Some of our discussion that was in isolation  
4 of interactive effects and we talked a little bit about it today  
5 and you may have some insights as we go through the charge  
6 questions, that perhaps while not directly related to what we do  
7 in the pesticide program, your report combined with the SETAC  
8 report any other reports, EFSA report, may help spawn research  
9 in different organizations in the federal government or  
10 internationally.

11 With regard to specific uncertainties that may  
12 relate to doing pesticide risk assessment, there is a variety of  
13 scenarios that could play out. The Part 158 test guidelines  
14 that some of the speakers discussed already has an opening -  
15 more than an opening, it has the ability to do different kind of  
16 field studies. But those guidelines are written, I think  
17 appropriately so, open and not very prescriptive because they  
18 are designed to answer specific questions.

19 Some of the feedback potentially from the  
20 charge questions could give us insights as to how to tailor, how  
21 to better define what the attributes of the semi-field studies  
22 or full-field studies, or maybe variations on the laboratory  
23 experiments could be to help focus, because our guidelines  
24 already give us some flexibility. That could be a way through  
25 chemical, by chemical or risk assessment scenario by scenario to  
26 get information, but you would be giving us some guidance on how



1 to be more efficient or effective in focusing the chemical risk  
2 assessment specific issues.

3 There could be some feedback that you provide  
4 that with the work we're doing with other countries and OECD or  
5 in other bilateral multilateral arrangements, there may be  
6 advice along with the SETAC Pellston document that may help  
7 advance some test guidelines, say at the laboratory level, that  
8 we all could use and then work with the regulating committee to  
9 go forward.

10 So, I think there is a variety of ways that  
11 this could play out. There may be some really pure research  
12 questions that we suggest and there are other parts of the  
13 federal family, state, international groups that will read your  
14 report and use it as well.

15 **DR. DANIEL SCHLENK:** Okay. Are we done? Any  
16 other questions? What we will do just to again summarize,  
17 mentioned this, this morning in our early morning, I think the  
18 Agency will be here tomorrow after our public commenting period.

19 Before we start questions, you'll have another opportunity to  
20 ask any questions that come out tonight in your discussions and  
21 deliberations, and just mulling over what has been gone over  
22 today that you ask one last time again before we actually get  
23 into the charge questions.

24 Okay. All right, before I turn it over to  
25 Fred, I just want to acknowledge thanks for the Agency's  
26 presentations today, and also for the panel if you would please



1 meet in the conference room after this, after you can do a  
2 restroom break, we need to meet and hash some things out before  
3 you head back to your rooms.

4 **DR. FRED JENKINS:** Okay. I just have a few  
5 things to say that are directed towards the public commenters  
6 for tomorrow. If you are giving a public comment that has a  
7 power point presentation, please see me as soon as possible,  
8 either now or early in the morning before the meeting,  
9 preferably no later than 8:30 tomorrow morning so I can load  
10 your power point presentation onto the computer behind me.

11 Also, to all the public commenters, please  
12 arrive to the meeting on time tomorrow at 9 a.m. We don't want  
13 you to miss your opportunity to provide your public comment.  
14 Once the public comment period closes, there will be no more  
15 opportunity to provide oral public comments at the meeting.

16 As a reminder, if you have not made prior  
17 arrangements with me to provide oral public comments, please see  
18 me as soon as possible so we can get you signed up as a public  
19 commenter and I ask also as a reminder, please limit your  
20 comment to five minutes.

21 And the last point I want to make, I ask that  
22 all public commenters please keep your oral comment times to the  
23 agreed upon time frame. If you've asked for five minutes,  
24 please only speak for five minutes, 10 minutes, please only  
25 speak for 10 minutes, an hour, please only speak for an hour.

26 We have a whole lot of ground to cover this



1 week, and we ask all of our public commenters to please help us  
2 in covering and accomplishing what we need to do for this  
3 meeting. Please stay within your specified timeframe. And  
4 that's all I have to say. This meeting is adjourned for today  
5 and we look forward to seeing everyone in the morning. Thank  
6 you for coming.

7 (WHEREUPON the meeting was adjourned for the  
8 day.)

9 **DR. DANIEL SCHLENK:** Okay. So, let's go  
10 ahead and just do a real brief introduction around the panel  
11 again. Just state your name and affiliations before we get  
12 started so everybody can identify themselves. My name is Dan  
13 Schlenk. I am professor in the Department of Environmental  
14 Sciences, University of California Riverside and I Chair of the  
15 SAP.

16 **DR. STEPHEN KLAIN:** My name is Steve Klaine.  
17 Good Morning. I am with Clemson University and I am an aquatic  
18 ecotoxicologist.

19 **DR. JAMES MCMANAMAN:** Good morning, I am Jim  
20 McManaman. I am at the University of Colorado in the Department  
21 of Obstetrics and Gynecology.

22 **DR. KENNETH DELCLOS:** Barry Delclos from the  
23 Food and Drug Administration's National Center for Toxicological  
24 Research.

25 **DR. DAVID TARP:** Good morning, I'm David  
26 Tarpy. I'm at North Carolina State University in the Department



1 of Entomology. I am a honey bee biologist and the extension  
2 apiculturist for North Carolina.

3 **DR. PAUL SCHWAB:** I am Paul Schwab from Texas  
4 A&M University. I am an environmental soil chemist.

5 **DR. THOMAS POTTER:** Tom Potter, USDA  
6 Agriculture Research Service, Southeast Watershed Laboratory in  
7 Tifton, Georgia. I am a pesticide chemist.

8 **MR. JENS PISTORIUS:** Jens Pistorius, Germany,  
9 Julius Kuehn Institute. I work with risk assessment for  
10 pesticides in honey bees, investigation of bee poisoning  
11 incidents and research pesticides in honey bees.

12 **DR. JEFF PETTIS:** Good morning, I am Jeff  
13 Pettis, I am with USDA Agricultural Research Service, Bee  
14 Research Laboratory here in Beltsville, Maryland, and I am an  
15 entomologist.

16 **DR. NANCY OSTIGUY:** Nancy Ostiguy, from  
17 Entomology Department at Penn State.

18 **DR. ROSALIND JAMES:** Rosalind James, I am with  
19 the USDA Agricultural Research Service. I am director of the  
20 Pollinating and Sex Research Laboratory in Logan, Utah.

21 **DR. GREG HUNT:** Good morning, I am Greg Hunt.  
22 I am at Purdue University and I study honey bee genomics,  
23 behavior and health issues.

24 **DR. NINA FEFFERMAN:** Good morning, I am Nina  
25 Fefferman from Rutgers University. I am an applied mathematical  
26 modeler of ecology and epidemiological systems.



1                   **DR. MAY BERENBAUM:** I am May Berenbaum, the  
2 Profession and Head of Entomology at the University of Illinois  
3 at Urbana-Champaign, and I am an insect chemical ecologist.

4                   **DR. DANIEL SCHLENK:** Thank you everyone.  
5 Before we get started, Fred has some comments he needs to make  
6 to our public commenters.

7                   **DR. FRED JENKINS:** Good morning everyone and  
8 welcome to the second day of our FIFRA SAP on Pollinator Risk  
9 Assessment Framework. My name is Fred Jenkins, and I am the  
10 Designated Federal Official for the FIFRA SAP. So, I just  
11 wanted to briefly say to the public commenters - we have quite a  
12 few public commenters this morning. If you have a power point  
13 presentation, please see someone on our staff so that we get  
14 your presentation loaded onto the computer behind us. Also, if  
15 you have any handouts of your comments that you wanted  
16 distributed to the Panel, please bring those now to our staff so  
17 that we can have those distributed as well. With that said, I  
18 am going to turn the floor over to Dr. Daniel Schlenk.

19                   **DR. DANIEL SCHLENK:** Thanks Fred. Couple  
20 comments -- this morning just speaking of materials - there were  
21 a couple papers that were placed in the chairs of some of the  
22 panel members. If you have materials that need to be presented  
23 to the panel they need to go through the EPA staff. We can't  
24 have materials distributed without a formal distribution pattern  
25 through the EPA. So again, if you have materials that you need  
26 to get to the Panel or you would like the Panel to see, please



1 do so through the EPA staff here. Okay. With that, we turn it  
2 over to the EPA and any sort of follow-up issues you need to  
3 present today before we start with the public comments.

4 **DR. DONALD BRADY:** No, thanks very much Dr.  
5 Schlenk, I think we are ready to go right to public comments.

6 **DR. DANIEL SCHLENK:** Great, what I would like  
7 to do, if you don't mind, is following the public comment period  
8 if you guys could come back up, I will ask the panel if they  
9 have any last minute questions related to the materials. Great,  
10 thanks.

11 Okay. Our first public commenter is CropLife  
12 America, and I believe that is going to be Dr. McAllister making  
13 that presentation. Again, as Fred had mentioned, please hold to  
14 the time that you have been allotted. We have an hour allocated  
15 for you guys, for CropLife, if you can please keep to that time  
16 that would be great.

17 **DR. RAYMOND MCALLISTER:** Thank you very much.  
18 My name is Ray McAllister. I am the Senior Director of  
19 Regulatory Policy for CropLife America. CLA is the National  
20 Trade Association representing the crop protectin industry in  
21 the United States. We would like to express our sincere  
22 gratitude to the bee technical workgroup, which has worked very  
23 hard over the past month or so to prepare the written comments,  
24 which have been distributed to you as well as the oral summary  
25 that you will be hearing this morning. I will just making a few  
26 introductory remarks, and then we will hear from David Fischer



1 of Bayer and Jay Overmyer of Syngenta for the bulk of the  
2 remarks today.

3 We all recognize that pollinator health is a  
4 vital issue for American agriculture. CLA in particular  
5 recognizes that crop protection technology, as well as the  
6 pollination services provided by both managed honey bee colonies  
7 as well as native pollinators are necessary for successful  
8 agriculture in feeding a hungry planet.

9 CropLife America commends the efforts of EPA,  
10 PMRA and Cal DPR in improving the science of pollinator risk  
11 assessment. Our member companies appreciate this opportunity to  
12 provide input into a very important process. We believe that  
13 improving the quality and rigor of risk assessments will ensure  
14 that the crop protection tools, which are critical for  
15 sustainable agriculture production, can be used in a manner  
16 compatible with pollinator health.

17 Pollinator health can be adversely affected by  
18 a combination of stress factors, which include a variety of  
19 diseases, some very devastating parasites, problem with  
20 nutrition, loss of habitat, chemical stress and bee management  
21 practices.

22 There are some who have asserted that  
23 pesticides are the root cause of colony collapse disorder in  
24 honey bees, but this is not supported by the weight of the  
25 evidence. We believe that establishment of a scientifically  
26 credible risk assessment approach for pollinators will help



1 promote sustainable agricultural practices and these are of  
2 benefit to all involved, from farmers, beekeepers and the  
3 American Consumer.

4 The establishment of sound and scientifically  
5 valid test methods and risk assessment processes should aid all  
6 interested researchers in optimizing study designs to ensure  
7 that the results are reproducible and that they can be  
8 interpreted clearly. Lack of such guidance has led to  
9 publication of some recent studies, which are not optimized for  
10 contribution to risk assessment and have led to some erroneous  
11 conclusions about the relative importance of pesticides in  
12 pollinator health in practical circumstances.

13 The improved risk assessments will be  
14 important to support the appropriate labeling of pesticide  
15 products and accompanying best management practices for  
16 safeguarding pollinator health.

17 CLA believes that the scientifically sound  
18 risk assessment procedures are critical not only to ensure that  
19 the EPA's and the nation's environmental protection goals are  
20 met, but to instill confidence in the public in the regulatory  
21 approval process. In the hopes of improving these risk  
22 assessment processes under review, we are offering the following  
23 comments, and I will turn the time over to Dave Fischer.

24 **DR. DAVID FISCHER:** Thanks Ray. I am David  
25 Fischer. I am an ecotoxicologist with Bayer CropScience, and on  
26 really the comments that I am presenting and Jay Overmyer from



1 Syngenta will be presenting is the work of - technical workgroup  
2 we have among the companies and CropLife America.

3 First, some general comments on the White  
4 Paper Risk Assessment Process. We support the harmonized  
5 approach among the three regulatory agencies. This is something  
6 that is of great benefit to our industry that we have a  
7 consistent approach stake in various jurisdictions. Not only in  
8 North America, but we would like to see harmonization around the  
9 world as well.

10 The tiered risk assessment approach we  
11 support. This is similar to what we do with other taxa. We try  
12 and screen out the chemicals that you can eliminate concern  
13 about, focus the resources where there is the main  
14 uncertainties; and we also support expanding the Tier I data  
15 requirements as the Pellston Workshop recommended and as the  
16 White Paper recommends to fill in some of the gaps we currently  
17 have with just the limited tests that are in the current  
18 guidelines.

19 We also support a flexible approach to doing  
20 higher tier testing, Tier II and Tier III. It would be nice if  
21 there was a prescription for doing this, but every chemical in a  
22 use pattern really will have its own differences that need to be  
23 taken into account. The key thing is to make sure that when we  
24 go to higher tiers, we are focusing on what the uncertainties  
25 are and addressing those with the studies.

26 We do have some concerns. We think the Tier I



1 assessment, especially the way the dietary exposure is being  
2 estimated, is overly conservative and will lead a lot of  
3 chemicals that are reasonably safe or really should not cause a  
4 concern progressing to higher tiers. I will say more details  
5 about that, why we think that might be true; and also how we  
6 think it might be remedied.

7 We are also concerned if we get into a system  
8 where a lot of chemicals have to go to higher tier testing that  
9 we have clear acceptability criteria in place for those higher  
10 tier tests and how they are going to be conducted, how they the  
11 data is going to be interpreted. Right now, there is a lot of  
12 uncertainty there.

13 So, we will go through sort of the parts of  
14 the White Paper with respect to problem formulation. We support  
15 the protection goals, which largely were the same as we  
16 developed at the Pellston Workshop, which our industry  
17 participated in along with the EPA scientists and academic  
18 scientists.

19 One thing that is appreciated in the White  
20 Paper that we have not always seen in other assessment documents  
21 is a real clear explanation for how levels of concern are  
22 established. We appreciate the clarity and the transparency of  
23 how they said this is what we are trying to do and this is the  
24 RQ value that seems to meet that criteria; and its science  
25 based, so we support that.

26 I mentioned flexible testing beyond Tier I. I



1 won't say anything more about that. Use of conceptual model -  
2 we thought that this White Paper has a very good illustration of  
3 how to do a conceptual model and how to use it to focus the  
4 assessment and the development of higher tier data to address  
5 the main uncertainties. It is very difficult to assess  
6 everything in an assessment, and I think this does a good job of  
7 focusing on that major routes of exposure.

8 Also, we support using honey bees as a  
9 practical surrogate for pollinators in general. This was  
10 discussed at length at the Pellston Workshop and the report will  
11 have some more information on non-Apis bees, but for practical  
12 purposes and certainly with the effects testing, the honey bee  
13 should be a reasonable surrogate.

14 Okay. Characterization of Contact Exposure -  
15 we basically support what is proposed in the White Paper using  
16 the relationship from the Koch and Weisser Database. It seems  
17 to be the best available data. It's consistent with what was  
18 agreed to at the Pellston Workshop. As the White Paper analysis  
19 shows, it seems to be an improvement on previous approaches.

20 Now into some areas where we see some room for  
21 improvement. Characterization of Dietary Exposure - first of  
22 all, the soil applications, the Briggs' model, we actually think  
23 is reasonably good Tier I starting point. You know, if you have  
24 to come up with an estimate of systemic uptake into plants, into  
25 field crops, this is a reasonable starting point. It doesn't  
26 necessarily tell you what is going to be expressed in the



1 flowers of a plant. It is not developed to do that, but it  
2 should be, if anything, an overestimate of that.

3 Again, as more data are developed, this will  
4 be a theme on a lot of our slides. Some of these Tier I  
5 estimates may need to be reevaluated.

6 With respect to seed treatments, the default  
7 value of PPM or mg/kg, I think was originally proposed for all  
8 systemic risk assessments. So, whether it was a seed treatment  
9 or a soil treatment, that seemed to be an upper-bound for seed  
10 treatments. However, the field data are quite a bit less. If  
11 you look at the field measurements in table 17, the highest  
12 value is 36 parts per billion and the Tier I estimate is more  
13 than an order of magnitude; it's like 30 times higher than that.

14 It looks to us like a default value of 0.1 mg/kg would be more  
15 appropriate and adequately conservative just based upon the data  
16 that we have.

17 For soil treatments, the numbers can be  
18 higher, but when they are the Briggs' model does an adequate job  
19 of reflecting that.

20 Again, I think maybe it's implied in the White  
21 Paper, but it doesn't specifically state, so we want to state  
22 it. There is no point in concluding there is a risk of use of  
23 seed treatments that aren't systemically active. There are a  
24 lot of them; especially a lot of the fungicides won't  
25 necessarily be expressed in the pollen and nectar to any extent.

26 We should be able to screen those out rather than just assume



1 that there is a concentration there.

2 Tree trunk applications - here is an area  
3 where we are in need of a lot more data, a lot of research. So,  
4 what the Agency has come up with is a back-of-the-envelope  
5 calculation that basically tries to say what if all the mass of  
6 compound applied is evenly distributed in the biomass of the  
7 tree, the leaves and flowers. There is a lot of uncertainty  
8 whether this is an accurate prediction. We think it is probably  
9 very conservative because flower concentrations are often orders  
10 of magnitude less than leaf concentrations, but it is a starting  
11 point.

12 So, you know, we think it is going to be very  
13 conservative. We do think that there may be some data out there  
14 from the USDA studies on Asian longhorn beetle control programs,  
15 Emerald Ash borer, maybe the Hemlock Woolly Adelgid. I think -  
16 and we don't have access to all these data, but there may be  
17 some data that USDA can pull in here that EPA hasn't considered  
18 yet.

19 Okay. So, our main concern is the foliar use,  
20 the dietary estimate that is being proposed is basically to use  
21 the tall grass value from the T-REX model, which really comes  
22 from the Hoeger and Kenaga nomogram as modified by Fletcher et  
23 al. For just looking at the numbers versus what these estimates  
24 are, the pollen numbers appear to be reasonably conservative.  
25 They're high. They're about 2X higher than the highest value  
26 that is in the dataset, but the dataset is small, so that



1 doesn't seem to be unreasonable as a starting point.

2 But for nectar, it's much, much higher. It  
3 appears to be overly conservative with the highest field  
4 measurements more than an order of two magnitude below what the  
5 T-REX model value is.

6 So, since nectar consumption, nectar  
7 concentrations really are what drive this risk assessment -- if  
8 you crunch the numbers 97 percent of the exposure comes from  
9 nectar and 3 percent from pollen according to the calculations  
10 -- it's critical that you not grossly overestimate the nectar  
11 concentrations.

12 So, just looking at the data, and I think I  
13 will transition to the next slide, looking at the data in table  
14 5, it looked to us that there is about a 3X factor between  
15 pollen and nectar, particularly when you look at the studies  
16 that are reporting average field measurements. We put more  
17 weight on those than the maximum ones because those can be just  
18 more variable.

19 So, we are proposing that you take the value  
20 for pollen and multiply it by 0.33 to get the estimate for  
21 nectar.

22 Then the question - how conservation should  
23 Tier I exposure estimates be? There is a statement that the  
24 Tier I method is intended to generate reasonably conservative  
25 estimates. We agree with that; but meaning that the estimates  
26 should generally be within one or two orders of magnitude higher



1 than the true environmental exposure. That seems to us to be  
2 too high. One to two orders of magnitude bias in your estimate  
3 can greatly change the number of the compounds that you fail to  
4 screen out and have to conduct higher tests with.

5 If you look at what Fletcher did in developing  
6 the nomogram values that are used in T-REX, they look at field  
7 residue measures and just took the 95th percentile. Really, if  
8 you look at EPA policy in the framework documents, they refer to  
9 high end estimates usually being in the 90th-95th percentile  
10 range; that is traditionally what is done. Now here we do not  
11 have a lot of data, so we may not have enough to just say use a  
12 95th percentile, but taking the highest value that you have and  
13 going 10X or 100X more than that seems to us to be too  
14 conservative.

15 We point out that the distributions and  
16 residue measurements tend to be log-normal, which means they are  
17 skewed to the low side with a fewer high-end values. Those  
18 high-end values can be statistical outliers and probably are  
19 less reliable. Sometimes, they may even be errors due to  
20 contamination when the sample is collected or analytic  
21 methodology.

22 Just what is the consequence if you over  
23 estimate exposure by 10 to 100 times? Just as an example, a lot  
24 of us probably had our morning coffee. If you look at caffeine,  
25 I don't know if you know this, but caffeine is actually pretty  
26 toxic.



1           The LD50 is in the range of 150-200 mg/kg body  
2 weight, which is about 80 cups of coffee with a typical strength  
3 of caffeine. So, that's about a little over 1 percent of an  
4 LD50 in each cup of coffee.

5           If you over estimate the caffeine  
6 concentration in coffee by 10X, you conclude that each cup has  
7 more than 10 percent of an LD50 dose, which in most risk  
8 assessment schemes would at least cause some concern. If you  
9 overestimate it by 100X, you can conclude that a cup of coffee  
10 is lethal to a lot of people. So clearly that would -- you know  
11 just as an analogy -- I think we would want to screen out very  
12 reasonably safe products like we would screen out having to do  
13 high risk assessments for caffeine in coffee.

14           The other thing is the food intake rate, which  
15 I think there is a lot of uncertainty, or at least we are  
16 wondering if that needs to be looked at - and I really  
17 appreciate what the Agency did because they pulled off a higher  
18 tier methodology approach by using a Monte Carlo procedure to  
19 try and take the data that Rortais et al had and come up with  
20 an estimate of how much food a bee ingests in a day based upon  
21 energetics and other things. We did a lot of the similar things  
22 in ECOFRAM a while back. Having done some of that modeling,  
23 some of the pitfalls were made aware to me because I made some  
24 of the same mistakes.

25           First of all, log-normal distribution was  
26 assumed for the sugar concentration in nectar. That might be



1 fine, but just setting the mean at 30 percent means that most of  
2 those trips out there, the assumption is that they're coming  
3 back with nectar with less than 30 percent because the  
4 log-normal is going to be skewed to the low side.

5 Of course, why is that significant? It's  
6 because you are basically totaling up the amount of nectar the  
7 bees are ingesting in order to get a certain amount of sugar and  
8 summing the mass of pesticide in that quantity of nectar. So,  
9 there could be some bias there. At least it is something that  
10 you might want to look at.

11 There is also an assumption that bees are  
12 foraging randomly. They don't show any preference for picking  
13 out food sources that have higher sugar content. I wonder if  
14 that is true. I would be surprised if it was. I think if you  
15 put a bunch of feeders out there, and there are probably people  
16 on the panel that know the answer to this, you would find that  
17 they would select the higher sugar source. You know, it would  
18 just be -- optimal foraging theory would predict they would.

19 There are three variables in this Monte Carlo  
20 Simulation that use uniform distribution. This is where some of  
21 my personal experience comes in. I did this myself. I put in  
22 uniform distributions in some of the ECOFRAM models and when  
23 Dwayne Moore reviewed them he told me, don't ever put in a  
24 uniform distribution. It puts too much weight on the tails,  
25 come up with some sort of beta distribution that uses expert  
26 judgement.



1                   So, I don't know if this changes - you know, I  
2 would want to see a sensitivity analysis to see if changing the  
3 distribution would change the result of the simulation.

4                   Then finally, number of foraging flights per  
5 day - I think and I could be wrong, but there was nothing that  
6 indicated to me when I read it that they weren't just assuming  
7 this was an independent variable from the distance traveled or  
8 the duration of the flight. It makes sense to me that wouldn't  
9 you expect this to be highly correlated that if you were making  
10 more flights per day, they were shorter flights. So, you  
11 wouldn't need as much energy, you wouldn't need as much sugar to  
12 fuel those flights.

13                  So, if you have correlated variables - and  
14 there are ways of doing that - you want to specify that in your  
15 modeling. I'm not sure if that was done. All of these factors  
16 may result in a nectar consumption being over estimated.

17                  So, I want to support the concept of using  
18 this sort of modeling technique. I just think it might need to  
19 be vetted.

20                  Dose from food intake - another thing is and  
21 this is common to assessments we do for birds and mammals as  
22 well - we do back-of-the-envelope, you know how many LD50s per  
23 day does an animal get? That's kind of what we are doing. We  
24 are calculating the cumulative dose over entire day of foraging  
25 per bee, but we are not taking into account metabolism and  
26 detoxification that is going on as this food is being ingested.



1 That can be a significant factor.

2           Going back to the caffeine example with  
3 coffee, you get a different dose if you have one cup of coffee  
4 an hour versus if you have 10 at once. You know, the caffeine  
5 is metabolized and detoxified, excreted so the doses don't  
6 necessarily add up, aren't completely additive. So, that is  
7 another source of conservatism in the assessment.

8           Then there are a lot of other factors. We  
9 support the option of addressing uncertainties in the risk  
10 assessment factors with whole colony exposure studies, which  
11 really is, I think, where we need to get better data and really  
12 find out what the colony is being exposed to.

13           Then there is in these other bullets, all  
14 these other factors that need to be taken into account as far as  
15 the use patterns and just how likely is exposure to occur. We  
16 would hate to see us wasting time doing higher tier assessments  
17 on exposure scenarios that should be screened out just on the  
18 basis that the way you are using a product as a bait or applying  
19 to a crop that bees aren't going to visit like leaf lettuce or  
20 applying post bloom. All of these factors I think EPA currently  
21 takes into account. We just want to emphasize that maybe the  
22 White Paper did not state that as clearly as needs to be stated.

23       So there will be other uses that will be screened out besides  
24 just based upon risk quotients.

25           Okay. I will turn the next piece on effects  
26 and risk assessment over the Jay Overmyer.



1                   **DR. JAY OVERMYER:** Thanks Dave. Again, my  
2 name is Jay Overmyer. I am an ecotoxicologist with Syngenta  
3 Crop Protection. I am going to go over a couple of the slides  
4 here related to effects testing.

5                   As Dave mentioned earlier, CLA supports the  
6 use of tier testing and the approach in the risk assessment.  
7 Obviously moving from Tier I to Tier III, if looking at the  
8 amount of ability to control the situation or control the test,  
9 there is a reduction in the amount of control from Tier I to  
10 Tier III. Also, moving from Tier I to Tier III, we also have an  
11 increase in environmental realism within those studies.

12                  So the Tier I studies are highly controlled  
13 laboratory tests designed to predict an LD50 or NOAEC, often  
14 subjecting the organisms to high doses to achieve those  
15 endpoints. Basically to come up with these endpoints, we are  
16 able to determine what the relative toxicities of these  
17 compounds are.

18                  Moving to Tier II, again a little bit less  
19 controlled, semi-field type studies. Exposure is closer to real  
20 world, but yet we are still looking at forced exposures with the  
21 bees inside of a tunnel or an enclosure, which inhibits them  
22 from naturally foraging that they might do. The benefits of the  
23 Tier II we're also increasing environmental realism, therefore  
24 we're getting up to a hive-level effect as opposed to  
25 individual-level effect.

26                  Then with Tier III, obviously again, we're



1     losing a little bit more control. The bees are allowed to  
2     forage naturally, often looking at natural application rates and  
3     exposures, and also being able to obtain information related to  
4     the hive, which is important.

5             For the Tier I testing, CLA does support that  
6     the use of the Tier I test, the adult contact and oral acute  
7     toxicity test as well as the seven day larval test when the  
8     methods are finally validated or for use of that. We also  
9     support development protocols for chronic toxicity testing of  
10    both adult bees and the larvae; and the industries interested in  
11    participating in this process if it is possible.

12            We also support the White Paper position that  
13    studies need to produce measured endpoints that can be clearly  
14    tied to assessment endpoints, such as colony strength and  
15    survival and the protection goals. We also feel that the  
16    internationally-common data requirements should have harmonized  
17    test guidelines so that we can use our studies for multiple  
18    countries and multiple regulatory agencies.

19            For Tier II, as Dave mentioned also that we  
20    like to see flexible study designs to be able to be produced to  
21    address uncertainties in the lower tier assessments; and we also  
22    realize that it is critical to communicate with the agencies on  
23    the study design such that we design studies that help answer  
24    questions that are apparent or has been useful for the risk  
25    assessment purpose.

26            I would also like to point out that within the



1 semi-field studies, again we are subjecting the bees to a  
2 somewhat unrealistic exposure scenario, again force feeding on a  
3 plot of a crop where insecticide has been applied or pesticide  
4 has been applied. And that in these studies they can produce  
5 false negatives or fail to show effects that occur at real field  
6 sites; but often produce false positives that show effects that  
7 don't occur at real field sites.

8 So again, this relates to the fact that we  
9 need to go to Tier III studies to help answer, again, some of  
10 these questions related to the uncertainty of Tier II.

11 So again, as with the Tier II studies, we also  
12 feel that it is necessary to consult with the agencies to  
13 develop appropriate protocols and site designs to help answer  
14 the questions that are necessary for risk assessment purposes.  
15 Studies should be designed and endpoints selected to provide the  
16 most appropriate information to assess protection goals.

17 Again, the study should be focused and help to  
18 answer specific questions rather than addressing all potential  
19 scenarios. Adding additional scenarios to study design creates  
20 more confusion as to how to interpret the results. So, keeping  
21 them focused would definitely be of benefit.

22 The Tier III studies incorporate potential  
23 effects of the plant protection products. It is multiple levels  
24 of organizations, so we would incorporate information from lower  
25 levels of biological organization up through the population  
26 level. So, we feel like this is, you know, important that we



1 focus on these higher level assessment endpoints that are able  
2 to relate to protection goals, such as protecting pollination  
3 services.

4 I would also like to point out in field  
5 studies, control plots are really reference plots because in  
6 most situations we have to apply chemicals to the control plots  
7 in order to grow a viable crop for appropriate comparisons. And  
8 also I would like to indicate that real-world biological systems  
9 are inherently variable and the variability should be properly  
10 interpreted within these studies.

11 The next two slides are a couple of graphs  
12 illustrating some data we obtained from some Tier III field  
13 studies. Again, these graphs are in the White Paper on page  
14 170.

15 The first graph here is illustrating effects  
16 of individual mortality, and that would be dead worker bees,  
17 pupae and larvae over the course of the study. As we see here,  
18 at specific days throughout the course of the study, we see  
19 differences in mortality between treatments and controls.

20 I would also like to point out that the  
21 highest level of mortality was observed prior to initiation of  
22 the treatment where the hives were brought into the actual study  
23 site.

24 Throughout the course of the study we do see  
25 again, variability within individual mortality. If we move to  
26 or look more closely at higher level assessment endpoint such as



1 colony strength -- and again this is estimated number of bees  
2 per hive -- we see that the controls and treatment track fairly  
3 well.

4 So again, at this higher level endpoint, it is  
5 incorporating all of the lower level assessment endpoints, the  
6 lower levels of biological organization into this higher level  
7 of biological organization endpoint. We feel that this colony  
8 level type of assessment is where we need to be as far as  
9 determining effects of potential compounds on the health of  
10 honey bees.

11 In reality, these assessment endpoints will  
12 then help looking toward protection goals. So, we are working  
13 towards determining endpoints that help to assess the protection  
14 goals as stated in the White Paper. With that, I will hand it  
15 back to Dave.

16 **DR. DAVE FISCHER:** Okay. So, in terms of risk  
17 assessment, again we support a tiered approach for all the  
18 reasons we have said. It is consistent with the standard  
19 methodology the EPA uses for other taxa.

20 Science-based approach to develop the proposed  
21 levels of concern, I mentioned that before; use of honey bees as  
22 surrogate for non-Apis bees; use of whole colony studies when  
23 triggered to put the results of laboratory testing with  
24 individual adult or larval bees into real-world context. This  
25 is an opportunity that we have with bees that we don't frankly  
26 have with some of the other species we do testing and risk



1 assessment for. It is hard to do a population level test with  
2 birds, fish and mammals.

3 Bees - that is what usually we want to protect  
4 is that level. With bees, we can test colonies and even acres.  
5 It is just an opportunity that we can test at that level that we  
6 don't always have.

7 Communication of the results of pollinator  
8 risk assessments should convey the uncertainties and inherent  
9 conservatism of the assessments methods. That is something we  
10 think is key with these Tier I assessments that are going to -  
11 there are compounds that are going to not be screened out and  
12 the concern is that that is going to be communicated as there is  
13 a risk. It doesn't necessarily mean there is any risk. It  
14 means that with the conservatism in the assessment, you couldn't  
15 say there wasn't one.

16 Population modeling - this is an area that a  
17 number of companies are investing in. We support the  
18 development of population modeling to address uncertainties in  
19 lower tier risk assessments. We are interested in participating  
20 in model development, and as I mentioned, we are investing some  
21 resources in that. Models should be transparent, have options  
22 for refinement and be well vetted before use by regulatory  
23 agencies.

24 So, to summarize sort of our technical review  
25 of this, there are a number of points of agreement. We think  
26 the new methodology represents an important advancement that



1 should improve the scientific credibility of pollinator  
2 assessments and increase the transparency and predictability of  
3 regulatory decisions. That's a good thing. We support the  
4 efforts of EPA, PMRA and CDPR to harmonize their pollinator risk  
5 assessments processes. Pollinator assessments should be based  
6 on scientifically sound methods and we think the proposal is  
7 consistent with that goal. Risk assessment should be  
8 interpreted within the risk-benefit context as required by  
9 FIFRA. That is particularly well explained in the White Paper.

11           There are some areas where we would suggest  
12 there are opportunities for improvement. Proposed screening  
13 criteria appear in some cases to be overly conservative and will  
14 produce many false positives. Many chemicals that pose no  
15 real-work risk to pollinators will fail the Tier I screen.  
16 There was some information presented on that, but I just thought  
17 I would throw up here that if you apply the Tier I dietary risk,  
18 it's basically - there is a fixed assumed dose per 1 pound per  
19 acre applied that is something like 32 micrograms that the bee  
20 ingests.

21           So, how high does your - I'm sorry I have to  
22 go back to my spread sheet - but basically to pass the  
23 assessment, you need an LD50 - if you have 1 pound per acre, you  
24 need to have an LD50 of 80 if you are applying 5 pounds per  
25 acre, the LD50 has to be above 400. We only test up to 100, so  
26 any chemical that is slightly above 1 pound per acre is not



1 going to pass this assessment, whether it is toxic or not unless  
2 we test higher. We think that the limit test should be enough.  
3 If you are down at 1/10 of a pound a.i. per acre, you need to  
4 have an LD50 higher than 8, which again that is a very low rate  
5 and that is not a particularly low LD50 number.

6 Just comparing it to the Hazard Quotient  
7 method in Europe - in Europe there is sort of a  
8 back-of-the-envelope calculation, which the application rate is  
9 compared to the LD50 and you compare that result, that Hazard  
10 Quotient to a trigger value of 50 and to pass that to get a  
11 Hazard Quotient that is less than 50 for 5 pound-per-acre  
12 application rate, the numbers are up there - 112 micrograms per  
13 bee is what the LD50 has to be for 1 pound per acre, 22, all of  
14 these are 3.6X different.

15 So this North American approach is 3.6X more  
16 conservative than the EU Hazard Quotient approach; and it's all  
17 because of the assumed dose that these are getting from nectar  
18 and the fact that you are estimating the nectar concentrations  
19 to be as high as they are.

20 And by the way I think everybody in Europe is  
21 pretty comfortable that Hazard Quotient is very conservative.  
22 We are going even more conservative with this one.

23 I mentioned that if you have a chemical that  
24 is nontoxic at the limit dose in the acute oral test then these  
25 chemicals should just be considered safe and you should not  
26 proceed to higher tiers.



1                   It is important the Tier I assessments are  
2                   used only to identify chemicals requiring further testing and  
3                   not for concluding or implying that such chemicals pose a  
4                   real-world risk, I mentioned that.

5                   Since higher tier testing will be triggered  
6                   for many chemicals, there must be clear guidance and achievable  
7                   acceptability criteria for higher tier studies. I mentioned  
8                   that at the outset. I will mention it again. That is a  
9                   concern. We have a long history of doing higher tier tests on  
10                  other taxa, for which there is a high rejection rate of the  
11                  studies when they are reviewed by the Agency. So, this is an  
12                  area where we see the need for a lot of discussion, maybe  
13                  workgroup, research, whatever and we are interested in  
14                  participating in the process.

15                  Just as a last slide, industry continues to be  
16                  proactive in advancing the science related to bee health issues  
17                  such as conducting research programs, developing best management  
18                  practices, formulation improvements, stewardship programs,  
19                  pollinator habitat initiatives, and this is an area that we are  
20                  active in and we want to continue to collaborate with the  
21                  agencies and with the research community to continue to advance  
22                  the science. Thank you for your attention.

23                  **DR. DANIEL SCHLENK:** Thank you for your  
24                  comments. Questions from the panel? Dr. Hunt? Dr. Hunt is  
25                  going to go first.

26                  **DR. GREG HUNT:** I wonder if given the problems



1 with Tier III testing and getting an acceptable study that EPA  
2 would accept, would CropLife be open to the idea if EPA only  
3 required Tier II testing?

4 **DR. DAVE FISCHER:** Jay might want to answer  
5 this as well, but I think we definitely want to have the option  
6 for field testing. At Tier II, we exaggerate the exposure when  
7 we do bee tunnel tests. We know of cases where there are  
8 affects in those tests that are basically false positives with  
9 respect to what happens in the field because the exposure is  
10 exaggerated. There is also the problem - I think the Agency  
11 gave a good summary of all the deficiencies or short comings of  
12 tunnel studies in that if you stress the bees you have limited  
13 amount of time that you can keep them.

14 So, I think we want to definitely have the  
15 option of doing full field testing and the challenge is to  
16 develop protocols where the results are clearly interpretable  
17 and everybody agrees on how to interpret the results.

18 **DR. GREG HUNT:** On the other hand, full field  
19 testings may lead to false negatives. I just wanted to make  
20 that comment.

21 **DR. DANIEL SCHLENK:** Thanks. Dr. Overmyer,  
22 did you want to add anything to that?

23 **DR. JAY OVERMYER:** No. I agree with what Dave  
24 had mentioned.

25 **DR. DANIEL SCHLENK:** Dr. James, you said you  
26 had a - no? Okay. Anyone else? Dr. Potter?



1                   **DR. THOMAS POTTER:** With regard to Tier II and  
2 Tier III, where does evaluation of mitigation practices fit into  
3 the framework and the approach?

4                   **DR. DAVE FISCHER:** We see mitigation as being  
5 an option at every stage of when we develop a product. Before  
6 we even submit it for a registration, we are determining what  
7 the hazards are to various taxa and developing a label with  
8 potential mitigation in mind; so some products, people like  
9 myself and Jay are asked by the company, do we need an at bloom  
10 restriction? Do we need to off label this crop or that crop  
11 because of particular attractiveness to pollinators.

12                   So, that is part of preparing the label and as  
13 you work through the system there should be an option for -  
14 instead of conducting another million dollars of work,  
15 mitigating the problem with a label restriction.

16                   **DR. DANIEL SCHLENK:** Yes, Dr. Pettis?

17                   **DR. JEFF PETTIS:** Yeah, Jeff Pettis. With  
18 regard to systemics and trying to predict concentration in  
19 nectar and pollen, do you feel that what is found in stems or in  
20 the plant tissue can accurately predict that? And also could  
21 you comment on just the ratio which we find in the few studies  
22 that we have about the concentrations in pollen versus nectar?

23                   **DR. JAY OVERMYER:** From the data that we  
24 generated, we typically see higher levels in the actual plant  
25 leaves and stems compared to what we see in nectar and pollen,  
26 but we do see variability within different plants. So, I think



1 it is important that we indicate that in generalization you  
2 cannot take one example and then extrapolate it to all different  
3 examples. As far as nectar and pollen goes, we typically do see  
4 more residues of pollen than nectar. Again, the ratios there  
5 vary between different plants.

6 **DR. DAVE FISCHER:** This is Dave Fischer. I  
7 will just add to that that it may differ between the chemistry,  
8 the compound involved. There are definitely a lot of chemicals.

9 Most of the systemic chemicals are largely xylem-limited in  
10 their transport in the plant. They will move with the water  
11 transport system, but not with the sugar transport system, but  
12 then there are some other chemicals that have the right chemical  
13 properties that will also be systemic in the phloem.

14 So, just looking at data and saying what is  
15 the concentration of stem versus the flowers, you really  
16 probably need to look a little bit more carefully about, you  
17 know how does that compare based upon the chemical properties  
18 that would affect their ability to move in these different  
19 transport systems. But as a general rule, the stem and leaf  
20 concentrations are an order of magnitude higher than what is in  
21 the flowers - what gets in the pollen and nectar.

22 **DR. DANIEL SCHLENK:** Okay. Any other  
23 questions? Okay. Thank you very much. Our next public  
24 commenter is CropLife Canada on my list. Presenter is Pierre  
25 Petelle.

26 **MR. PIERRE PETELLE:** Good morning. So, yeah,



1 my name is Pierre Petelle. I am the Vice President of Crop  
2 Protection Chemistry at CropLife Canada. I just have a few  
3 slides to do through, and you will see probably a lot of  
4 repetition from what we just talked about because we do  
5 collaborate quite a bit with our colleagues to the south here.

6 Our technical working group in Canada, we  
7 actually share some members with the technical team here at CLA.

8 We collaborate on this file in particular because it is a very  
9 important file for the industry. CropLife Canada and our  
10 members support the CLA position and the comments you just heard  
11 on the White Paper.

12 As indicated previously, pollinators and  
13 pesticides are complementary components of the sustainable  
14 agricultural system. Many of the crops grown in Canada, like  
15 here in the U.S., depend on both crop protection and pollination  
16 services.

17 So, a sound scientific approach to  
18 understanding impact of pesticides on pollinators is going to be  
19 absolutely critical here.

20 As Ray pointed out earlier, bee health is a  
21 complex issue. Our industry is committed to the protection and  
22 promotion of pollinator health, and we are very supportive of  
23 efforts to advance the science. So, as we move forward in this  
24 process, we want to reiterate the need for scientifically sound  
25 risk assessment procedures and the use of data from  
26 well-designed, valid scientific studies.



1 Canada and the U.S. enjoy a long history of  
2 cooperation and collaboration in the pesticide regulatory area.  
3 PMRA and EPA in particular have been world leaders in the area  
4 of regulatory harmonization, pesticide reviews, undertaking  
5 joint reviews, work sharing projects and just improving the  
6 scientific processes. I think this has helped both growers and  
7 regulators and our industry on both sides of the border in terms  
8 of access to newer safer products, in terms of using the most  
9 modern science and in terms of efficient regulatory review.  
10 This process has been developing over the past 15 years at  
11 least, and you know, I think as we move forward with this  
12 particular issue, we are highly supportive of harmonized data  
13 requirements, test guidelines and risk assessment processes. We  
14 are glad to see the role that PMRA has played in this whole  
15 process with the White Paper and here at this Panel.

16 So just lastly, this is a reiteration of some  
17 of the comments that CLA made a few minutes ago, but in order to  
18 ensure the protection goals are met and instilling public  
19 confidence, we circle back to this need for a scientifically  
20 sound approach to risk assessment. We feel that the proposed  
21 approach is broadly consistent with this goal looking forward.

22 In particular, we want to stress the  
23 importance of the tiered risk assessment approach that was just  
24 discussed in detail, and the use of honey bees as a good  
25 surrogate for non-Apis.

26 That's all I wanted to say today. Thanks very



1 much, and we again appreciate the role of PMRA in this process  
2 and stress the importance of scientifically rigorous process as  
3 we move forward.

4 **DR. DANIEL SCHLENK:** Any questions from the  
5 panel? Dr. Berenbaum?

6 **DR. MAY BERENBAUM:** Given the differences that  
7 exist in apicultural practices in Canada and the U.S. and  
8 variance on restraints, do you think standards are completely  
9 harmonizable, or do you think that there still may be means for  
10 recognition of differences?

11 **MR. PIERRE PETELLE:** Yeah, I mean I think in  
12 terms of the science and the risk assessment process, I think  
13 there is an ability to harmonize those processes. Are there  
14 Canadian specific or U.S. specific practices that need to play  
15 into that? I mean that is going to have to be a question as we  
16 move forward, but from our experience up to this date,  
17 harmonization of those processes should be fine.

18 **DR. DANIEL SCHLENK:** Thank you very much.

19 **MR. PIERRE PETELLE:** Thank you.

20 **DR. DANIEL SCHLENK:** Our next presenter is the  
21 Center for Regulatory Effectiveness, and that is Mr. William  
22 Kelly.

23 **MR. WILLIAM KELLY:** I do not have any slides  
24 or handouts. My name is Bill Kelly. I am with the Center for  
25 Regulatory Effectiveness. We are an independent organization  
26 that evaluates regulatory issues that are of widespread concern



1 and usually have quite a bit of controversy. We are also  
2 placing great emphasis on what we call information quality and  
3 the Information Quality Act and its guidelines.

4 We have already submitted some detailed  
5 written comments, lots of footnotes and quotations, so I won't  
6 bother you with that.

7 Dr. Steeger yesterday at the end of the  
8 meeting emphasized that the high quality of EPA studies and the  
9 studies that are conducted or required to be submitted under  
10 their guidelines, and we recognize that. I think that the  
11 problem we have is with a lot of journal articles.

12 The Information Quality Act Guidelines  
13 recognize that peer review of journal articles is of very uneven  
14 quality. A lot of articles are published simply because they  
15 are, let's say, thought provoking and not really contributory to  
16 the knowledge base. Those need to be examined carefully for  
17 compliance with the standards and guidelines under the IQA.

18 Near the end of yesterday's meeting, the Panel  
19 asked a good question that relates to the Information Quality,  
20 and that is what to do with data gaps.

21 Data gaps are not really a problem with Tier I  
22 or Tier II, but when you get to the final risk assessment and  
23 you have gone through Tier III, the way the data gaps have to be  
24 handled is a matter of law under the IQA and its guidelines is  
25 for the scientists -- that are the risk assessors as opposed to  
26 the risk managers -- to simply describe the data gap and its



1 significance, and then leave it to the risk managers to decided  
2 what to do with that. The data gap should not be filled with  
3 assumptions or default values or whatever. Those are usually  
4 policy-driven values that should not be used in the risk  
5 assessment process.

6 Another good example that came up very late  
7 yesterday after unfortunately a lot of people had left the  
8 meeting was the issue of what we are really dealing with here.  
9 The extent and I would say also the distribution of the  
10 pollinator decline, which is often equated with colony collapse  
11 disorder unfortunately. A chart was presented, which showed the  
12 decline was presented by Mr. Steiger or Mr. Carrington. Then we  
13 found out that that chart was based on honey production. So,  
14 that would not pass muster under the Information Quality Act  
15 Guidelines for example.

16 I mean we really do need good data on the  
17 extent of this problem and its distribution. I think that that  
18 is important, especially for Tier I, especially the  
19 distributional aspect of it. I mean if you look at distribution  
20 of the problem and the extent of the problem in various areas,  
21 you might find out that there are say, crop areas. We're not  
22 talking about states necessarily, but crop areas or particular  
23 crops where there is a high level of decline, but a particular  
24 pesticide is not used on those crops or in that area. That  
25 could be a basis for eliminating that pesticide from Tier I.

26 On the other hand, you could have an area in



1 which a particular pesticide is used very intensively, and yet  
2 there is no significant increase in pollinator decline beyond  
3 what you would expect from, you know, say ordinary winter kill.  
4 So again, that could factor into Tier I determinations.

5 This issue of the extent of pollinator decline  
6 and the distribution of the problem is really missing from the  
7 framework at this point. There could be an interagency  
8 coordination issue here because it appears that USDA has taken  
9 the lead on this issue.

10 Back in January, the Inspector General of the  
11 Department of Agriculture issued a report that was on progress  
12 the USDA had made in evaluating this issue. The quote from the  
13 report says, "The true extent and impact of CCD -- which they  
14 equated with general decline -- in the United States has not  
15 been adequately assessed despite USDA's use of significant  
16 resources for honey bee research to address CCD."

17 The EPA is part of that steering committee  
18 that is working on this issue. And as I understand it, part of  
19 the problem is money either from Congress or with redirecting  
20 resources within the agencies.

21 So, I think in looking at the matter of  
22 problem formulation, this issue is really missing from the  
23 framework at this point and needs to be added. The SAP needs to  
24 address it. We need to do some sort of pattern analysis on  
25 where this problem is occurring, the extent of it and whether it  
26 is really associated with pesticide use in those areas.



1 And finally, I think if we are going to do  
2 this since it is missing from the framework at this point, we  
3 need some further public comment on this particular aspect of  
4 the issue. Okay. That is all I have at this point. Are there  
5 any questions?

6 **DR. DANIEL SCHLENK:** Thanks. Dr. Berenbaum?

7 **DR. MAY BERENBAUM:** Actually, I am a little  
8 confused. In view of the fact that bees are livestock and  
9 there's at least some legal precedence that bees are owed the  
10 duty of common humanity if they trespass on agricultural land,  
11 why is documenting decline relevant to risk assessment of  
12 pesticide use on agricultural land?

13 **MR. WILLIAM KELLY:** Thank you. Well,  
14 presumably this framework and this risk assessment could lead to  
15 FIFRA Regulatory Action, particularly on registration. FIFRA,  
16 as a matter of law, requires a balancing of risks and benefits.  
17 In order to do that, you are going to need to quantify the risks  
18 together with the benefits, and you can't do that with some  
19 accurate appreciation of the extent of the impact of what's  
20 occurring. Does that.

21 **DR. DANIEL SCHLENK:** Dr. Berenbaum?

22 **DR. MAY BERENBAUM:** Well as you pointed out,  
23 bee decline is not equal to bee mortality. So, bee death per se  
24 isn't colony collapse disorder, but still risk assessment for  
25 pesticide use involves determining bee susceptibility, bee  
26 mortality. So, I am still not sure where the linkage to



1 documenting decline, at least for honey bees.

2 **MR. WILLIAM KELLY:** I think what we are  
3 interested in is decline. I don't know how we would measure  
4 decline in health short of colony loss and bee mortality. At  
5 least that's how the problem has been posed to date in terms of  
6 colony loss and mortality. If we want to start looking for sick  
7 bees, I guess that takes us to another level.

8 **DR. DANIEL SCHLENK:** Okay. Any other  
9 questions or comments? Okay. Thank you very much. Our next  
10 presenter is Scott Schertz with the National Agricultural  
11 Aviation Association.

12 **MR. SCOTT SCHERTZ:** Hello. I do appreciate  
13 the opportunity to be in front of the SAP on the pollinator  
14 protection issues, and we do support this initiative.

15 Basically, well my name is Scott Schertz. I  
16 do own and operate Schertz Aerial Service, an aerial application  
17 operation in central Illinois in corn-soy-bean country. I am a  
18 past president of the NAAA and the comments have to do with  
19 basically aerial application of crop protection products.

20 The NAAA does represent a majority of the  
21 aerial application industry in the U.S. There are approximately  
22 1700 members in 46 states, and the majority of the members of  
23 the NAAA are operations such as mine or small or medium-sized  
24 businesses that provide these services to many crops, everywhere  
25 from the major crops to many of the specialty crops.

26 As the White Paper reflects that mode of



1 application is one of the areas of concern, you know I would  
2 like to give a little introduction to the aerial application  
3 industry as far as our scope and qualifications.

4 Aerial application does account for  
5 approximately 25 percent of the application of crop protection  
6 products in the U.S. and commercial bases, and in many areas, it  
7 is the preferred ways of applying due to its speed, able to get  
8 over tall crops or in conditions that make ground application  
9 impractical.

10 In addition, that means that aerial  
11 application does have, you know, a unique opportunity on  
12 pollination protection in that the airplanes and helicopters can  
13 apply a lot of product very rapidly and in a narrow time window.

14 When the hives - the areas of the prime bee activity are known  
15 as many states required, this does allow a tiered approach,  
16 basically applying at time of low pollinator activity.

17 I do want to emphasize though, that that  
18 doesn't mean that it is practical for all applications to be  
19 done then; and that knowledge of where they are is very, very  
20 important as I will emphasize many states require.

21 When you start looking at the crop protection  
22 needs of, you know, many of the major field crops that I am very  
23 familiar with, there is a need for viable products with  
24 reasonable label requirements, even during pollination time. I  
25 mean, that is when the majority of the insects actually affect  
26 plants, and obviously there is a conflict here in times.



1                   And then also our members do serve many of the  
2 specialty crops that may be intensively utilizing managed  
3 pollinators, and in those cases, I mean these types of things of  
4 spraying at night are common. But it doesn't, I mean, it isn't  
5 a matter of just getting more airplanes. I mean, this is a very  
6 specialized industry. There are a finite number of airplanes.  
7 The airplanes are very sophisticated. There is a lot of  
8 technology in them that I will go through briefly in a couple  
9 minutes.

10                  Then also the night operation is a very  
11 specialized subpart of it. Yet, some areas it is common;  
12 typically it is in more of the specialty crop areas. In my  
13 area, yes I do some night work, but there are also many fields  
14 that it is impractical and unsafe to do at night. When you  
15 start talking about dealing with obstacles such as met towers,  
16 winter events, large power lines, the judgement at night, the  
17 depth perception is a real issue. So, you can't just say well  
18 that is going to magically fix all of these issues.

19                  The industry is very active on refining the  
20 safety programs and the deposition of the airplanes. Our main  
21 program is something called PASS (phonetic) that has been in  
22 effect for about 14 years. It is something that is an active  
23 training session that goes around the country and is available  
24 to the entire industry. Basically, the subjects have to do with  
25 drift mitigation, aeronautical decision making, security and  
26 pollinator issues have been something that has been addressed in



1 the last several years.

2 Another unique thing about the aerial  
3 application industry is what is called our safe program.  
4 Obviously, these details are in the paper submitted, and that is  
5 a program that is also unique to aerial application industry on  
6 an infield way of measuring the deposition from the airplanes  
7 and refining that to be appropriate for the products to be used.  
8

9 So, I did want to give you a bit of  
10 introduction and how that, you know, aerial application does  
11 have some unique opportunities on this, but you will also need  
12 to be realistic that it isn't a silver bullet of just doing  
13 everything at night.

14 There is further detail in the paper that was  
15 submitted that you should have a copy of. I do appreciate that  
16 opportunity to be in front of you. Any questions?

17 **DR. DANIEL SCHLENK:** Any questions from the  
18 Panel? Okay. Thank you Mr. Schertz.

19 **MR. SCOTT SCHERTZ:** Thank you.

20 **DR. DANIEL SCHLENK:** Our next presenter is  
21 James Doan, who is an owner of a bee keeping operation.

22 **UNIDENTIFIED SPEAKER:** Mr. Doan is running  
23 late.

24 **DR. DANIEL SCHLENK:** Okay. Is Mr. Jenkins  
25 available, for Center for Food Safety?

26 **MR. PETER JENKINS:** Greetings. I am Peter



1 Jenkins, Attorney and Consultant working with the Center for  
2 Food Safety. You should have copies of the oral comments that I  
3 am going to give you now.

4 We are in contact with Mr. Doan, and he is  
5 hung up on the Metro somewhere. Hopefully he will be here  
6 before the end of the comment period. Dr. Jenkins if you could  
7 be a bit lenient on that, and we will let you know when he shows  
8 up. He is a beekeeper who came from New York.

9 Let's see, I am just going to basically read  
10 my comments. We have written comments that are in the record  
11 and I will be glad to answer any questions at the end about  
12 either the oral or written comments.

13 I am pleased to make these comments on the  
14 White Paper that was submitted in support of the risk assessment  
15 process or the proposed RA process for short.

16 My organization, the Center for Food Safety,  
17 has got comments in your packet. Please give those your close  
18 attention because our comments as well as the comments of Beyond  
19 Pesticides and some of the other NGOs involved here -- and  
20 beekeepers -- have submitted some detailed scientific studies  
21 that are omitted from the proposed RA process document that we  
22 think very much relate to this process.

23 Let me stress the critical role you are  
24 playing at ensuring a state-of-the art risk assessment framework  
25 to protect honey bees and native bees from excessive and perhaps  
26 unsustainable mortality from pesticides. It appears,



1 unfortunately, that EPA lacks inhouse PhD levels entomological  
2 expertise, at least very much of it that we can see. So, you  
3 can help to provide that.

4 The process you are a part of has involved  
5 real world cost and impacts already. Years of delay and  
6 thousands of staff hours have gone into creating the proposed  
7 framework, but it still has major areas of uncertainty and  
8 omission. Further, EPA has refused to follow through on the  
9 prior condition that had it required years ago when it  
10 registered two neonicotinoid insecticides, Clothianidin and  
11 Thiamethoxam, for which providing a valid bee impact field  
12 study, a pollinator field study or Tier III sort of test as we  
13 have been hearing about, was made a condition to continue in the  
14 registration of those pesticides by EPA. That condition has  
15 still not been met nine years after it was first imposed in the  
16 case of Clothianidin.

17 An excuse that EPA now uses is its condition  
18 currently is being "held in reserve" pending the outcome of this  
19 FIFRA SAP process. So you, as SAP Panelists, have become part  
20 of EPA's non-precautionary approach to this issue of pollinator  
21 field test design for these pending conditional registrations.

22 So, now you can at least help make sure that  
23 the cross bar on the shifted goal post is high enough to be  
24 really comprehensively protective. It has taken so long to get  
25 to this point.

26 A written comment highlighted the inadequacy



1 of the proposed RA framework as far as taking sublethal effects  
2 into account. Our comment also urged modeling for both the  
3 contaminated dust and the guttation fluid exposure routes for  
4 systemic neonicotinoid insecticides, and to have a clear call  
5 out in the RA framework for more Tier II and Tier III testing,  
6 if necessary, of those routes before approving such insecticides  
7 in the future.

8 On the neonicotinoid contaminated dust route,  
9 it was very inappropriate for the RA framework on page 99 to  
10 include evasive statements from the agency that in effect say,  
11 the SAP can just rely on EPA to mitigate the risks of this route  
12 through discussions with seed treatment registrants and the  
13 manufacturers of seed planting equipment.

14 That sort of vague, hopeful, self-serving  
15 assertion restated at least twice by EPA officials yesterday has  
16 no place in a risk assessment framework.

17 EPA has no real power to enforce such a  
18 speculative, undefined requirement for modifying tens of  
19 thousands of planting machines across the entire country, and  
20 even if EPA had such power, the practical obstacles to EPA  
21 enforcing that sort of requirement in any reasonable timeframe  
22 far exceed the Agency capabilities.

23 This contaminated dust exposure route and its  
24 impact on honey bees has led to Germany, Italy, Slovenia, to  
25 some extent France, prohibiting various neonicotinoid seed  
26 treatments in the last four years. So relying in EPA's attempts



1 to downplay this route in the U.S. would be irresponsible.

2 The SAP must demand a more rigorous treatment  
3 of this route or it won't be doing its job. Please take careful  
4 account on this issue of the written comments by Dr. Christian  
5 Krupke of Purdue University that in the packets - I hope they  
6 are in your packets - which reinforce what I have just said.

7 Proposed RA framework document makes  
8 occasional mention of EPA's ecological incident information  
9 system or EIIS. I would remind the SAP members that it is well  
10 accepted across numerous stake holders, especially beekeepers,  
11 that the EIIS does a very poor job of collecting national bee  
12 kill incident data.

13 In contrast, Canada's PRMA reporting system is  
14 well regarded. Here is an illustration. In EPA's July 27, 2012  
15 response letter to a petition filed by my organization, the  
16 Center for Food Safety, 25 beekeepers and other NGOs to suspend  
17 the registration of Clothianidin, the Agency stated with respect  
18 with EIIS, "We are however aware of 14 additional incidents  
19 occurring in the U.S. in 2012 that are not yet present in the  
20 database and approximately 120 additional incidents reported in  
21 Canada."

22 Extensive media and non-EIIS reports make  
23 clear that both Canadian and U.S. beekeepers suffered vast  
24 numbers of spring 2012 bee kills due to the contaminated dust  
25 exposure route associated with corn planting just this past  
26 spring. It is not plausible at all that 120 documented bee kill



1 incidents associated with neonicotinoid seed treatments  
2 involving several thousand bee colonies occurred in Canada's  
3 relatively small corn planting area -- mostly in southwestern  
4 Ontario and Quebec as we understand it -- but only 14 additional  
5 bee kill incidents occurred in the entire U.S. across its orders  
6 of magnitude larger corn areas.

7 The seed treatment products, machinery used  
8 and environmental conditions do not change dramatically when you  
9 cross the border. What changes is the reliability of the bee  
10 kill reporting systems. My point here is that a revised RA  
11 framework should not rely on the EPA, EIIS unless it is  
12 dramatically improved.

13 On the issue of drinking water - our written  
14 comments focused on the guttation liquid route and those are  
15 important comments, but I urge your attention to a separate  
16 exposure route that our written comments did not really address.

17 A key paper by Starner and Goh 2012, is Detections of the  
18 Neonicotinoid Insecticide, Imidacloprid, in Surface Waters of  
19 Three Agricultural Regions in California. It documents that a  
20 significant portion of sampled surface waters were contaminated  
21 above EPA allowed levels for chronic invertebrate exposure  
22 across diverse agricultural landscapes.

23 These waters are wide-spread sources of  
24 drinking water for all bees, but the proposed RA framework does  
25 not mention this California DPR paper, nor does it quantify the  
26 risk to bees from the real world levels that were measured in



1 that study. Instead, it follows a very questionable academic  
2 exercise in appendix 2 in the RA framework to discount this  
3 drinking water route all together.

4 The fact this environmental contamination by a  
5 major neonicotinoid exists now in California and elsewhere is  
6 indicative of Agency failure to present undue consequences and  
7 it has passed inadequate risk assessments. The SAP can now  
8 correct this failing going forward.

9 On the portion of the document addressing the  
10 risks to non-Apis bees, I think any fair appraisal of the  
11 framework document would conclude that this is its weakest  
12 section. After years of delay, merely suggesting that there  
13 might be future "modifications" of the basic RA framework to  
14 address the unique circumstances of thousands of native North  
15 American bee species is utterly inadequate to the risks that you  
16 are facing here.

17 Given that many native species have small  
18 localized ranges, the RA process must at least explicitly assess  
19 the need to restrict or limit the use of pesticides in these  
20 limited locations, which is a common sense way to reduce risk to  
21 these localized species. Several of these non-robust species  
22 already are disappearing and lack any margin for Agency error  
23 moving forward and ensuring their survival.

24 To the extent that the proposed RA framework  
25 has overlooked, down played or minimize various exposure routes,  
26 the classes of sublethal effects, the existing U.S. National Bee



1 Kill Baseline and the other issues such as cumulative and  
2 synergistic impacts that I can't go into, the document  
3 constitutes a best case scenario from the perspective of the  
4 companies that you have heard from seeking to approve  
5 pesticides. But it is a worst-case scenario from the  
6 perspective of bee conservation and preservation and preserving  
7 beekeeper livelihoods.

8           You hold the intellectual key to finding the  
9 right medium between these best and worst case scenarios. If  
10 you fumble an important element of the risk framework, you could  
11 put the long-term sustainability of these species, particularly  
12 the thousands of relatively poorly studied native bee species in  
13 jeopardy.

14           I want to hit one last point because it just  
15 came up in a previous talk by CropLife. I have been looking at  
16 the RA framework on page 170. There are two figures, figures 12  
17 and 13; and some generalizable point is made about those  
18 figures. It is in our written comment. We said what are the  
19 labels, what is the citation, what is going on in these figures  
20 12 and 13, because they are basically cleansed of any  
21 information except to try to make a generalized point.

22           Well, it turns out that those figures are the  
23 same ones that the gentleman from Syngenta showed in his slides.  
24 He actually had labels on his slides indicating what was going  
25 on there.

26           So, it appears that EPA has just taken



1 Syngenta's figures, put them into the RA framework at page 170  
2 without the labels and tried to make a generalizable point about  
3 it, but the point doesn't flow and it doesn't make sense. So, I  
4 just bring that to your attention. It's a bit concerning that  
5 EPA has used an unattributed bit of information from Syngenta or  
6 CropLife, whoever it was that put it together and just stuck it  
7 into the document.

8 So, with that, again I am an attorney. I  
9 cannot answer scientific questions, but I am glad to answer any  
10 questions that I can. Thank you.

11 **DR. DANIEL SCHLENK:** Thank you for your  
12 comments. Any questions from the Panel? Okay. So let's go  
13 ahead and take a break right now - 15 minutes - be back at  
14 10:45. We will continue with our public comments at that time.

15  
16 (Brief recess.)

17 Okay. If I can have everybody take their  
18 seats please. Okay. Is James Doan here? Yes. Okay. Mr.  
19 Doan, if you don't mind, we are going to have you come second  
20 after Beyond Pesticides comes forward if that's okay. Sorry  
21 about that.

22 So, can I have Nichelle Harriott, is she in  
23 the room? As Nichelle is coming forward, just to point out, I  
24 believe Dr. Fefferman you had asked for the OECD Guidance  
25 Document on the Validation of Structure Activity Relationship  
26 Models? That is on the docket. So, for anybody else who would



1 like to do that, just go onto the website and you can down load  
2 a copy of that. Okay. Ms. Harriott?

3 **MS. NICHELLE HARRIOTT:** Hi, good morning.  
4 Thank you for allowing me to present these comments. My name is  
5 Nichelle Harriott. I am representing Beyond Pesticides. Beyond  
6 Pesticides is a not-for-profit organization that works at the  
7 grassroots level to protect public and environmental health,  
8 identifying the hazards of chemical-intensive land, building and  
9 community management practices, and promoting healthy,  
10 sustainable and organic systems.

11 Beyond Pesticides is very concerned about the  
12 status of our nation's pollinators, especially the honey bee.  
13 Each year since 2006, commercial beekeepers have reported  
14 unprecedented bee losses. Normal losses occur around 17.6  
15 percent, but over the 2008 and 2009 period an estimated 29  
16 percent of all U.S. bee colonies died.

17 Since the honey bee is the most economically  
18 valuable pollinator worldwide, responsible for the pollination  
19 of high value crops such as almonds and broccoli, it is  
20 important that these pollinators are protected from the  
21 potential harmful effects of pesticides under EPA's  
22 jurisdiction.

23 One class of pesticides in particular has  
24 emerged as a major suspect in recent bee decline. Several  
25 studies have demonstrate that neonicotinoid insecticides like  
26 Imidacloprid, Clothianidin and Thiamethoxam have sublethal



1 effects in honey bees, which include disruptions in mobility,  
2 navigation and feeding behavior. Lethal and sublethal exposures  
3 have been shown to decrease foraging activity along with  
4 olfactory learning performance and decreased hive activity.

5 When exposed to sublethal doses of  
6 neonicotinoids at levels present in the environment, honey bees  
7 were less likely to return to the hive after foraging. Bees  
8 exposed to these chemicals also exhibit convulsions,  
9 uncoordinated movements and tremors that are typical of  
10 neurotoxic insecticide exposure. One study analysis of dead or  
11 dying bees by Krupke et al. found residues present at the range  
12 of 3.8 to 13.3 parts per billion.

13 A study by Schneider et al. shows that under  
14 field-like conditions, as little as 0.05 to 2 nanogram per bee,  
15 Clothianidin can significantly reduce foraging activity. In  
16 context, Reetz et al. found that corn seeds treated with  
17 Clothianidin result in translocated concentrations up to 8000  
18 nanograms per milliliter. For the honey bee, toxicity has been  
19 observed at an LD50 of 22 nanograms per bee for Clothianidin, 13  
20 nanograms per bee for Thiamethoxam; levels lower than those  
21 noted in EPA's Clothianidin registration documents, which report  
22 an LD50 more than 43.9 nanograms per bee.

23 In fact Clothianidin and Thiamethoxam are only  
24 second in toxicity to Imidacloprid with a LD50 17.9 nanograms  
25 per bee.

26 Studies have also reported that bees exposed



1 to sublethal doses of pesticides are highly susceptible to  
2 pathogens that lead to their decline. A 2012 study by USDA  
3 researchers discovered that newly emerged bees exposed to  
4 sublethal levels of Imidacloprid during larval development and  
5 indirectly from brood food from nurse bees had higher levels of  
6 the gut parasite Nosema, which is known to adversely affect  
7 colony health.

8 Systemic uses on corn seeds means that  
9 pesticide residues are translocated throughout the plant to  
10 pollen and nectar. Pesticide-intensive corn cultivation, which  
11 accounts for 80 million acres of land planted in the U.S.

12 According to Krupke et al., the application  
13 rates for Clothianidin on corn can range from 0.25 to 1.25 mg  
14 per kernel. A single kernel therefore contains several orders  
15 of magnitude of active ingredient more than the published LD50  
16 values for honey bees. The translocation of residues throughout  
17 the plant means that pollen and nectar are highly contaminated.

18  
19 Similarly, Clothianidin has been found in or  
20 on nearby plants like dandelions resulting from translocation of  
21 residues from the soil to the flower, from surface contamination  
22 of the flowers from airborne particles, or from a combination of  
23 both factors.

24 The panel, when making its recommendations to  
25 the EPA, must always keep in mind that bees are exposed to  
26 chemical residues through multiple routes of exposures. Bees



1 are exposed to pesticides via foliar and systemic treatments  
2 when they are pollinating flowering crops, and pesticide drift  
3 from surrounding areas.

4 They are also exposed to residues from wild  
5 plants contaminated through cross-pollination with corn  
6 systemically treated with pesticide. Bees are also exposed to  
7 residues in plant exhaust material produced during the planting  
8 of treated seed. Guttated water of seed-treated plants, which  
9 provides a source of water for bees, is also a source of  
10 contamination and exposure.

11 Similarly, bees can be exposed to multiple  
12 pesticides at any one time while foraging. It is now accepted  
13 and supported by the scientific literature, that chemical  
14 mixtures can pose greater health hazards in combination. In  
15 fact, research by scientists at the University of Florida linked  
16 bee decline to larval exposure to a cocktail of pesticides.  
17 Unfortunately, EPA's risk assessment process does not  
18 accommodate chemical mixtures, which means that synergistic  
19 effects continue to go unevaluated.

20 Pollinator field studies must therefore  
21 reflect real world events. Study designs should be based on the  
22 planting practices of the crop treated with pesticide, including  
23 evaluating the impacts of farm equipment that may result in  
24 airborne residues.

25 Field sites must be large enough to  
26 accommodate the distances travelled by foraging bees, and hive



entrances should be placed in a location to ensure bees will sample the field site. Life-cycle studies should also be a requirement. It is also important to note that non-Apis bees, in addition to pollen and nectar, are exposed to soil and tree pesticide residues.

EPA states that the protection goals for honey bees include protecting pollination services, honey production and bee diversity, with the endpoints being acute lethality, survival, growth and the reproduction of the colony.

For years, the science has shown that exposure to pesticides is detrimental to bees, and that levels not immediately lethal to worker adults may cause significant hidden damage to colonies. We hope the panel places an emphasis on a reliance on broadly representative, multiyear, controlled field tests, to provide realistic data points from which to judge a pesticide's impacts on honey bees.

It is imperative that the EPA has available all the relevant data before any pesticide product can be registered and introduced into the environment. It is unacceptable that EPA allows pesticides to be sold and distributed, or expands uses of existing registrations without knowing how these pesticides would impact bees, other pollinators and essential nontarget organisms.

In closing, we wish to impress upon the panel the importance for finalizing a uniform, comprehensive protocol that aligns with the pollinator protection goals stated by the



1 Agency.

2 Again, thank you for this opportunity to  
3 comment and Beyond Pesticides has already submitted written  
4 comments to the docket with a list of relevant scientific  
5 studies for the Panel's consideration. Thank you.

6 **DR. DANIEL SCHLENK:** Thanks for your comments.

7 Any questions from the panel? Okay. Thank you very much.  
8 Okay. Mr. Doan, we will get you next here.

9 **MR. JAMES DOAN:** Good morning. My name is  
10 James Doan, and I live in Hamlin, New York. I own and operate  
11 Doan Family Farms, an apiary where we keep 2000 hives of honey  
12 bees. Our apiary places 1600 hives in fruit orchards in the  
13 spring and we place another 1000 hives in vegetable crops in the  
14 summer. Our hives travel south to Fort Meade, Florida for the  
15 winter, and we place our bees in orange groves in the spring for  
16 orange honey production.

17 Beekeeping has been in my family for 57 years.  
18 Myself - I have kept bees for 44 years. My son, Ben, has also  
19 recently joined the family business and will be a fourth  
20 generation beekeeper. I come here today to talk about a problem  
21 that we have seen this year in our industry and that I feel must  
22 be fixed.

23 In May of this year 2012, I saw our apiary's  
24 first heavy bee kill since 1987. Our honey bees that were hit  
25 then were with a chemical called methyl parathion. The  
26 Department of Environmental Conservation that year documented



1 and did tests on the hives showing the very high loss at that  
2 point due to methyl parathion. What we saw this year was very  
3 much like methyl parathion. We haven't seen a bill kill like  
4 this since that point 25 years ago, and I really don't want to  
5 see damage like this again.

6 We had placed bees in an apple pollination  
7 location in Red Jacket, Geneva in March of this year. We were  
8 called to remove them, and when we got there this spring, the  
9 bees were dead, crawling on the ground in front of the hives.  
10 These are huge hives of bees, single stories, nine frames a  
11 brood, two boxes of honey on them.

12 We removed them, took them back to my farm. I  
13 called the DC, which is our Department of Conservation that next  
14 morning. They refused to come out. The lady at Region 8  
15 Department of Environmental Conservation told me they had no  
16 budgetary money to come and look at any bee kills. I will side  
17 note here - if I shot a bear, they would have been out in about  
18 15 minutes. Bees are not a priority apparently, in New York, to  
19 be tested.

20 So I called our New York State Inspection  
21 Service. The New York State Inspection Service said, yeah we  
22 will come out, at least we can check for mites, we can check for  
23 nosema, and we will pull samples, because we knew at this point  
24 we had to do something to follow through. We had a good live  
25 bee kill. The bees were still coming out the next morning just  
26 piling out of these hives.



1                   When he got there, the bee inspector, Bob  
2                   Duncan, opened the hives up. They had two boxes of honey capped  
3                   off on them. There was eight to nine frames a brood in a  
4                   single-story colony bees there were about two to three frames of  
5                   bees left at that point.

6                   He collected up wiggling bees on the fronts of  
7                   the hives and in the grass in front of the hives, put them into  
8                   bags, froze them, and he took them to his place and held them.

9                   At that point, we didn't know quite where to  
10                  send them. So, I called Maryann Frazier down to Penn State.  
11                  She said that they had a program where they would pay half the  
12                  cost of getting the samples run. We sent them to her. She sent  
13                  them on to the USDA bee lab down in Gastonia.

14                  They ran them through as quickly as possible  
15                  because, as I said, we've never seen anything like this at the  
16                  same time -- and you will see, I have an illustration, there is  
17                  a picture here on the forth page. That is what the bees looked  
18                  like the day we got them back to our place out front. That is  
19                  after already they have been at least one or two days into bee  
20                  kill on that.

21                  I called the farmer the next morning also. I  
22                  wanted to find out what the hell he had done to my bees. He  
23                  said he had not sprayed yet. They were waiting for us to get  
24                  the bees out. The only thing he told us was the guys around him  
25                  were planting corn at that point.

26                  So, we took that information, you know, along



1 with pulling the samples and that. What we came back with is as  
2 you can see on my next page is they checked for mites of course.

3 Our mite levels range from 9 to 3 to 18. They weren't  
4 extremely high, high enough where we probably had been in  
5 treating, but we wouldn't have to worry about that.

6 Nosema samples - the first sample they pulled  
7 came back negative, so there wasn't nosema on that one. The  
8 second one came back with 3,600,000 spores, which is still, in  
9 the bee industry today, is not considered a high level of nosema  
10 anymore. I know men that are running 50 to 75 million spore  
11 counts and still not suffering huge losses.

12 We got back our results and on the page  
13 following the nosema report, you will see what things were found  
14 in the pollen. The first page is the pollen. You know, and  
15 certainly some of the things we found, Clothianidin was one of  
16 them, Captan in the pollen at extremely high levels, but in the  
17 bees themselves, Clothianidin was the thing that showed up.

18 I mean, there are a few other things here, but  
19 nothing that really strikes you like Clothianidin with all the  
20 bees sitting there in those orchards with corn being planted  
21 around them.

22 So, you can take your assumptions for what  
23 they are as far as how the bees got into Clothianidin. It was  
24 an apple orchard with dandelions. They shouldn't have been out  
25 working in any corn fields at that point, and I don't know if  
26 they had been flying through dust. But that brings us to the



1 next problem.

2 We lost all those hives, all 148.  
3 Consequently, we tried to put splits and nukes back into those.  
4 Those all died also. So, it was useless for us to waste our  
5 time in doing that. We might as well burn the equipment and  
6 that. That is an economic huge loss to me, 148 hives out of  
7 2000 is a lot of money.

8 The next thing I want to show you is in June  
9 on the last page, you will see a big color photo, and those are  
10 mostly all drones. I have about 47 bee arches that had an inch  
11 thick pile of drones a foot in front of the hives. I haven't  
12 gotten the test results back, but I can tell you that is not  
13 nosema and its not rural mites.

14 So, I don't know what chemical it is, but I  
15 have never seen it. I can tell you that I've talked to guys  
16 across the country, nobody has ever seen this. The genitalia  
17 are out on these bees, some of these drones are only days old.  
18 This is not normal. It's something that you need to be aware  
19 of. There are chemicals out here that are doing really bad  
20 things in bee arches.

21 Consequently what happened after this was we  
22 had no matings on our queens, so all the hives that swarmed -  
23 drone layers or queen-less hives. My loss this summer is  
24 running over 20 percent and that is after we put in 1000 queens  
25 on a 2000 hive operation over the summer. This is pretty  
26 dramatic stuff.



1           You know, I didn't put it in here, but we are  
2 probably re-queening 100 percent of our hives over the course of  
3 a year, and I'm not alone in what is going on. It's everyone  
4 replacing huge amounts of queens in their operations just to  
5 maintain the hives they have right now. And we are still losing  
6 bees.

7           I mean, don't get me wrong, there are other  
8 problems out there, but when I see bees laying in front of hives  
9 like this, that is not those problems.

10          You know, I know there is a big problem - I  
11 want to go back to some of the other problems I see are the LD50  
12 of Clothianidin. I don't know who runs the test exactly beside  
13 the applicant, but in the tests that they do on LD50, do they  
14 specify the age of the bees they are working with? You know,  
15 contact versus oral - we've got so many multitudes of variables  
16 with these things. I am not sure the LD50 - I think it needs to  
17 be re-looked at.

18          Then we talk about synergy, stacking products.

19          You know, I talked to one of my corn friends, he has seven  
20 different products that go on his corn seed when they go in the  
21 ground. What does that do? I mean, when guys are spraying  
22 apple orchards, they don't just put in one product. They are  
23 mixing two or three at the same time. What's that doing and  
24 what do those things together? Is that being looked at?

25          You know, another issue is the tolerance  
26 level. I don't know how many of you are aware of it, but there



1 are only three products that have a tolerance level at honey,  
2 fluvalinate, coumaphos and fenpromexite. That's it.

3 All this other stuff that we find in bee  
4 hives, it isn't supposed to be there; and I can tell you today  
5 the push is in the United States, they want pollen and honey in  
6 those containers of honey. They want beeswax. We know what's  
7 in beeswax guys and ladies. We know what's in pollen. I mean,  
8 this isn't rocket science.

9 I mean, one of these days, there's gonna be a  
10 big issue, who's is going to pay for that cleanup and mess?  
11 What I am going to do with barrels of honey that I can't do  
12 anything with? I can't sell them to anybody and I have to take  
13 them to the HAZMAT? No. We need tolerance levels for all of  
14 these chemicals that are out there today in honey. Captan, you  
15 know, all of them. We just have to have them. You have them in  
16 every other food out there. We have to have them.

17 You know, EPA's job is to protect the citizens  
18 of this country from unregulated pollution in our environment.  
19 As a citizen, I don't feel I'm protected right now. I have  
20 honey bees that are being killed by products that are not  
21 supposed to kill bees. I have pesticides that cause my cost to  
22 go up the tree. And because the results are increased disease  
23 levels, they threaten my livelihood. War has been started over  
24 less and I feel as though I'm in one; a war to save the bee and  
25 me.

26 Please look at the way data I gathered on



1 incidents. Please look at LD50s, please work on getting  
2 tolerance levels in honey for chemicals, and please help us save  
3 the bees. Thank you.

4 **DR. DANIEL SCHLENK:** Thanks for your comments.  
5 Any questions for Mr. Doan? Okay. Thanks a lot. Appreciate  
6 it. Oh you have one, Dr. Pistorius?

7 **MR. JENS PISTORIUS:** Yes, Pistorius. I was  
8 wondering, maybe I missed it during the talk. You said that one  
9 incident where you provided pictures that a lot of drones were  
10 found dead.

11 **MR. JAMES DOAN:** Right.

12 **MR. JENS PISTORIOUS:** Was that the colony that  
13 the relation of female bees to drones were changed and do you  
14 think that a lot of drones were affected only?

15 **MR. JAMES DOAN:** It seemed like only drones.  
16 In that picture, there are some workers, but predominantly it  
17 was drones -- all the drones in the hives and every hive it was  
18 like that.

19 **MR. JENS PISTORIOUS:** Okay, thank you.

20 **DR. DANIEL SCHLENK:** Dr. Fefferman?

21 **DR. NINA FEFFERMAN:** Sorry, this is just  
22 scientific curiosity, do you have any impression from that?  
23 Were the drones trying to get back in and being forced out, or  
24 were they just flying out?

25 **MR. JAMES DOAN:** No, we were in a major honey  
26 flow at that point. Those drones were not kicked out. They



1 were crawling in the grass in front of the hives dying. We are  
2 still waiting on the results back from that. I have 50 bags of  
3 samples to run. We have lots of samples collected. It is a  
4 matter of who can pay for all these to be run. Right now,  
5 beekeepers are paying the cost of running all these things. Yes  
6 sir?

7 **DR. GREG HUNT:** So, in those same hives  
8 earlier in the year, you had a bee kill.

9 **MR. JAMES DOAN:** Right.

10 **DR. GREG HUNT:** And now this is June.

11 **MR. JAMES DOAN:** Right.

12 **DR. GREG HUNT:** Okay. I just wanted to get  
13 the background.

14 **MR. JAMES DOAN:** Now those 148 hives  
15 completing were died after the second time. We just push that  
16 stuff off to the side and dump all the frames out and burned it  
17 up. I couldn't - I was - I couldn't deal with them. There was  
18 nothing there - I wasn't going to put anymore bees into it.

19 **DR. GREG HUNT:** So these drones were in other  
20 hives?

21 **MR. JAMES DOAN:** Yes sir, in every yard, every  
22 hive in every yard.

23 **DR. GREG HUNT:** In every yard. The crops  
24 around were similar?

25 **MR. JAMES DOAN:** Well, our area is  
26 predominantly corn, soybeans, cabbage, apples, it has dairy,



1 it's a mixture of everything.

2 **DR. GREG HUNT:** Okay. Thanks.

3 **DR. DANIEL SCHLENK:** Dr. Berenbaum?

4 **DR. MAY BERENBAUM:** Did you have to pay for  
5 the Gastonia analysis?

6 **MR. JAMES DOAN:** Yes.

7 **DR. DANIEL SCHLENK:** Dr. Pettis?

8 **DR. JEFF PETTIS:** Back to the incident  
9 reporting, I realize that state by state, there's a lot of  
10 variation in that. The EPA, I believe, has set up a way of  
11 directly reporting, have you tried that at all?

12 **MR. JAMES DOAN:** I did do that once I found  
13 out about it, but there were a lot of beekeepers in New York  
14 state that didn't know about it at the time and a lot of  
15 beekeepers didn't have samples pulled. I've talked to people  
16 from all across the country that had very difficult time trying  
17 to get somebody from their state to come up and pull samples. I  
18 have talked to the Canadian beekeepers and they're telling me  
19 very similar stories to what I'm telling you, other than they  
20 got a lot of help out of their ministry to pull samples. Thank  
21 you very much.

22 **DR. DANIEL SCHLENK:** Any other questions?  
23 Okay. Our next presenter is Dr. Mike Beevers from California  
24 Agriculture Research.

25 **DR. MIKE BEEVERS:** Can you hear me? Yeah, my  
26 name is Mike Beevers. I'm a research director for California



1 Agricultural Research, and we're a contract research  
2 organization sometimes called a private lab. We've done  
3 regulatory honey bee studies for about 26 years. I'm just here  
4 to share a little bit of perspective of the people who are doing  
5 some of the work right now and may be doing a lot of the  
6 research in the future, just to let you know kind of our  
7 understanding of this.

8 The subject here is Potential Changes in  
9 Regulatory Research Requirements for Pollinators. Regulatory  
10 Research is the bailiwick of the contract research organization.

11 We perform scientific studies for submittal to EPA or similar  
12 government entities by manufacturers of agricultural products.  
13 Our goal is to provide independence, objectivity and reliability  
14 in the studies.

15 Our studies produce data to answer very  
16 specific risk assessment concerns regarding pesticides held by  
17 government regulatory bodies and our data is confidential. I am  
18 trying to just describe this as regulatory research.

19 Exploratory research is something entirely different and seeks  
20 to answer more broad ranging hypothetical questions. Most of  
21 this work is peer reviewed and is not under GLP and it is done  
22 at the universities. Obviously, this work is very important  
23 going forward with bees, the university work especially.

24 Contract research organizations assure the  
25 public of their independence by following good laboratory  
26 practices as established by the EPA.



1 I was going to ask the Panel, are you familiar  
2 with the Good Laboratory Practice Provisions? Just to let you  
3 know, we do function under these. These standards are the  
4 cornerstone of most contract research organizations, and there  
5 are specific standards regarding the data, the recording of  
6 data, raw data, protocols, study director, legal  
7 responsibilities, quality assurance, in-life audits, data  
8 auditing of final reports, archiving, standard operating  
9 procedures, short-notice inspections by the enforcement branch  
10 of the EPA out of Denver.

11 The purpose of these guidelines is to assure  
12 that the agencies, when they get data from our labs, are getting  
13 something they can rely on that they can go back years later and  
14 produced to defend their decisions.

15 A very simple example of GLP Practice would be  
16 placing a sample of nectar or pollen in a freezer. That freezer  
17 has to have a maintenance log. The sample must be logged into  
18 the freezer. The freezer must have 24/7 temperature monitoring.

19 It has to have alarms, probably a generator backup for power  
20 failures. When the sample is removed, it must be logged out,  
21 chain of custody, so forth and so on. I am just trying to  
22 indicate that there is a lot of work that goes into these  
23 studies to make them valid.

24 Noncompliance with these laws can result in  
25 fines and imprisonment. There are people on contract labs who  
26 have actually gone to prison. I probably don't need to explain



1 that if working under these circumstances, people take the data  
2 very, very seriously. I just would like to communicate that to  
3 you guys, and I appreciate you listening on this.

4 The lab's interest is in alignment with the  
5 Agency's interest in terms of data quality and objectivity. The  
6 people in the labs are interested in learning the newest methods  
7 and learning to implement them.

8 I would just like to say that, you know, the  
9 people in these labs, and I'm speaking for myself, would like to  
10 be a resource in the future as part of this and we are operating  
11 under a very strict guideline.

12 Finally, if anyone knows of any problems with  
13 labs -- some of these labs have been criticized in the past -- I  
14 would be interested in talking with them at some point. That is  
15 all I have to say, thank you.

16 **DR. DANIEL SCHLENK:** Thank you. Any questions  
17 for Dr. Beevers? Dr. Pettis?

18 **DR. JEFF PETTIS:** There has been a lot of work  
19 in Tier I testing on *Apis mellifera*. Have you done any testing  
20 with other species of pollinators at all?

21 **DR. MIKE BEEVERS:** Not successfully, no. The  
22 non-*Apis* issue is very tricky for, as I understand it, for  
23 getting predictable, replicable tests. The honey bee is just a  
24 wonderful organism to work with in comparison with those.

25 **DR. JEFF PETTIS:** I wouldn't argue about the  
26 honey bee, but my concern is that if we were to move to other



1 species testing, what would be reliable?

2 **DR. MIKE BEEVERS:** We would be - I have worked  
3 with leaf cutter bees. We have tried starting with bumble bees.  
4 Yes, that can be done and we are interested in doing it, but to  
5 be very honest with you, I haven't had a lot of demand for that.  
6 But that is an area we are interested in, and we would love to  
7 try.

8 **DR. DANIEL SCHLENK:** Okay. Any other  
9 questions? Yep, Dr. Tarpy?

10 **DR. DAVID TARPY:** This is Dave Tarpy, NC State  
11 University. Is there an analogous kind of GLP, but not for the  
12 L but for kind of like ecological setting that would hold up to  
13 that same stringency as the GLPs that you work under?

14 **DR. MIKE BEEVERS:** That's an excellent  
15 question, and not to my knowledge. The studies in ecological  
16 settings are extremely complicated. We have applied the good  
17 laboratory practice to the data. There're a lot of things that  
18 would transfer there, but in terms of actually addressing some  
19 of the possible issues that you may have in mind, we are dealing  
20 with a good laboratory practice for that purpose right now.

21 **DR. DAVID TARPY:** Thank you.

22 **DR. DANIEL SCHLENK:** Any other questions? Dr.  
23 Pistorius?

24 **MR. JENS PISTORIUS:** Assuming you have a lot  
25 of contact with other GLP laboratories in the U.S. too, my  
26 question is have you established in vitro test method yet as a



1 routine method and with acceptable control mortality, and are  
2 you able to do to tests longer than day 7?

3 **DR. MIKE BEEVERS:** The answer to that is  
4 essentially no. The in vitro test has been tested -- no from me  
5 personally. Some of the other labs may have developed that, but  
6 I'm not aware of. We have ring tested one in vitro test, but my  
7 understanding is it's still in development.

8 **DR. DANIEL SCHLENK:** Any other questions?  
9 Okay. Thank you, Dr. Beevers.

10 **DR. MIKE BEEVERS:** Thank you.

11 **DR. DANIEL SCHLENK:** Our next presenter is Rod  
12 Snyder from the National Corn Growers Association.

13 **MR. ROD SNYDER:** Good morning. My name is Rod  
14 Snyder, Director of Public Policy for The National Corn Growers.  
15 We appreciate the opportunity to just have a few brief remarks  
16 this morning. I will be providing comments on behalf of NCGA,  
17 which represents more than 37,000 members in 48 states and more  
18 than 300,000 corn farmers who contribute to state checkoff  
19 programs across the country.

20 I want to focus just these very brief  
21 statements on the benefits of seed treatments since that is  
22 where we've been getting a lot of questions recently regarding  
23 the studies for their potential impacts on pollinators.

24 Seed treatments are critically important to  
25 growers because they protect the seeds from disease and pests  
26 from planting through germination. Unpredictable weather



1 patterns during the planting season could lead to the seed  
2 remaining in the ground for an extended period of time and  
3 without these products, yields would be reduced and entire  
4 fields may need to be replanted.

5 Seed treatments lead to a higher yield corn  
6 crop by providing a healthier root system, particularly by  
7 maximizing early season plant stands. Growers also consider  
8 these seed treatments an important investment as they seek to  
9 protect and maximized high-value seed.

10 Field tests for the products have demonstrated  
11 a 6 bushel per acre average yield increase in corn. For  
12 example, Clothianidin is an important neonicotinoid seed  
13 treatment for corn in the U.S. The product is used in  
14 approximately 60 percent of corn planted in this county. It is  
15 a valuable product as growers seek to control wireworms, black  
16 cutworms, white grubs and other early-season pests that attack  
17 corn seeds and seedlings at a period when they are most  
18 vulnerable.

19 It is also worth noting that in no-till  
20 practices, no-till operations, because the soil health is  
21 improved, it also means additional pest pressures. So, all of  
22 the benefits from no-till operations could be lost if we have to  
23 start tilling again because of pressure on the seeds.

24 The use of seed treatments reduces the  
25 potential human health effects for corn growers because it  
26 reduces the need for soil-applied sprays or granules.



1 Environmental exposures are also reduced due to the fact that  
2 the seed is incorporated beneath the soil surface at planting  
3 time. This leads to a lower probability of exposure to birds or  
4 runoff to water bodies when compared to soil-applied products.  
5 Seed treatments actually have the effect of reducing the amount  
6 of active ingredient needing for effective pest control.

7 Eliminating foliar applications also removes  
8 the concern with making extra trips across the field during a  
9 time of the year when rainfall and muddy soils can create major  
10 time constraints during planting season. Fewer trips across the  
11 field also leads to fewer greenhouse gas emissions from fossil  
12 fuels used in tractors and other farm equipment.

13 While corn growers remained concerned about  
14 the recent decline in bee populations in the U.S. and the  
15 potential impact on the larger agricultural economy, studies  
16 suggest that there are multiple and complex causes of colony  
17 collapse disorder that cannot be explained by a single class of  
18 chemicals or agricultural practices.

19 There are no demonstrated long-term effects on  
20 bee colonies from neonicotinoid pesticides. Furthermore, corn  
21 growers work diligently to minimize dust from planters and often  
22 establish relationships with nearby beekeepers to minimize  
23 exposure to pollinators. We appreciate the opportunity to  
24 comment today, and we urge this Panel to maintain a  
25 scientifically sound risk assessment process when reviewing the  
26 impact of seed treatments on pollinators. I am certainly happy



1 to follow up with other information that you need. Thank you.

2 **DR. DANIEL SCHLENK:** Any questions? Yes, Dr.  
3 Hunt?

4 **DR. GREG HUNT:** Yeah, I've heard that  
5 undoubtedly seed treatments are good for corn yields. I've  
6 heard the 6-bushel an acre data before. Is that in a published  
7 study, and has there been a study that looked at the benefit of  
8 just the neonicotinoids without the other fungicides, et cetera?

9  
10 **MR. ROD SNYDER:** Sure, I'd have to go back and  
11 figure as we were piecing this together where that citation came  
12 from, but we can easily follow up on that. Yeah, absolutely.

13 **DR. DANIEL SCHLENK:** Dr. Potter and Dr.  
14 Berenbaum?

15 **DR. THOMAS POTTER:** Yeah, I just feel  
16 compelled to make a comment here because regarding your point  
17 that you made about increased pest pressure with no-till  
18 practices associated with improved soil health. I'm not sure  
19 what you mean by that, but I think there're probably other  
20 issues other than improved soil health that may cause increased  
21 problems with pests, probably just the fact that the soils are  
22 colder and wetter.

23 I don't mean to make too big a point about  
24 that, but I think improved soil health is a really good thing.  
25 It is something that I've connected my career mostly to, so you  
26 know, I think there are other issues than soil health that may



1 contribute to pest pressure in no-till systems, which certainly  
2 are important conservation system set up throughout the country.

3 **MR. ROD SNYDER:** Yes, by all means. Improved  
4 soil health is a big part of the reason why farmers shift to  
5 no-till, and I'm sorry if I've left any confusion there. There  
6 are many, many benefits to no-till systems. Our growers are  
7 moving in that direction rapidly in the last 10 or 20 years.  
8 But you are right. Whether it is the temperature of the soil of  
9 what have you, there can be additional pest pressures in those  
10 settings. So, seed treatments allow that adaptation to no-till.  
11 That's just, sorry, the basic point I was trying to make.

12 **DR. MAY BERENBAUM:** Wireworms and white grubs  
13 and black cutworms are not uniformly distributed across the  
14 corn-growing region and differ in their importance as economics  
15 pests. Do you know if there is - that 60 percent of corn  
16 growers who use pretreated seed - is that predominantly in  
17 regions where there is history of pest problems?

18 **MR. ROD SNYDER:** As far as the distribution of  
19 where the product is used, I'd have to look that up and get back  
20 to you. I mean certainly it's a common product in the corn-belt  
21 in the midwest and upper midwest, but I can look that up for  
22 sure.

23 **DR. DANIEL SCHLENK:** Any other questions? Dr.  
24 Pistorius?

25 **MR. JENS PISTORIUS:** Short one. How high do  
26 you think is the percentage of non-tillage systems for maize



1 cropping?

2 **MR. ROD SYNDER:** I'm sorry, the question is  
3 how -

4 **MR. JENS PISTORIUS:** How big is the portions  
5 of fields where there is no tillage for sowing?

6 **MR. ROD SNYDER:** That is a little bit region -  
7 there are regional differences. I can say with certainty in the  
8 mid Atlantic region where there's been a lot of emphasis on  
9 water quality in trying to prevent runoff because of the  
10 Chesapeake Bay situation, recent studies have shown that close  
11 to 90 percent of acreage in this region has moved to either  
12 no-till or conservation tillage. It is a little lower in the  
13 midwest, but I mean, it's a very high percentage.

14 **DR. DANIEL SCHLENK:** Dr. Hunt?

15 **Dr. GREG HUNT:** Just real quick, I was a  
16 little surprised by you saying that only 60 percent of the  
17 growers are using treated seed.

18 **MR. ROD SNYDER:** That was Clothianidin  
19 specifically. Other products would drive that number much  
20 higher. Clothianidin has a lot - there has been a lot of talk  
21 about that product in particular.

22 **DR. GREG HUNT:** Perhaps up to 98 percent.

23 **MR. ROD SNYDER:** Right.

24 **DR. DANIEL SCHLENK:** Okay. Any other  
25 questions? Okay.

26 **MR. ROD SNYDER:** All right, thanks.



1                   **DR. DANIEL SCHLENK:** Thank you. Our last  
2 public commenter today is Stephen McFadden from Independent  
3 Scientific Research Associates. And Mr. McFadden, if I could  
4 have you to about a 5 to 10 minute range, is that appropriate?  
5 Thanks.

6                   **MR. STEPHEN MCFADDEN:** Hello. I'm from  
7 Midland, Texas and after all the academic discussion, I want to  
8 point out where the rubber hits to road. We recently had a  
9 larger urban aerial spraying in Dallas, Texas. The DFW  
10 Metroplex is 6.5 million people. It's the largest metropolitan  
11 area in the south. The city of Dallas is 300 square miles. In  
12 2003, we beat the West Nile virus using an IPM program put  
13 together by former EPA'er, Bill Curry.

14                   The West Nile virus virulence has a feedback  
15 term where if you have a bad year, the next year a lot of birds  
16 are immune. And so, you have a good year the next year. But  
17 after nine years, then the birds aren't immune.

18                   So, after nobody was watching the store for  
19 2010 and 2011, in 2012 we had the worst outbreak probably in the  
20 U.S. ever. They aerially sprayed 1150 square miles twice,  
21 mostly two nights in a row, homes of maybe 2 million people.  
22 They used Clarke Duet with Sumithrin and Prallethrin from  
23 twin-engine planes at 300 feet at night. From 300 feet, the  
24 spray probably takes between 20 minutes and 2 hours to hit the  
25 ground, I'm guessing. So there is probably a lot of drift.

26                   A few organic farmers were upset. Some



1 beekeepers were upset. I would image that a number of bees were  
2 upset.

3 There were presumably a bunch of governmental  
4 affairs, three issues. One of our issues is like on the City of  
5 Dallas website it says that aerial spraying is safe and  
6 effective. EPA does not allow the term safe to be used to apply  
7 to pesticides, but this came from the top state health office in  
8 Texas who was using the term safe including on a CDC  
9 teleconference, and nobody confronted him on it.

10 The remedy is to sticker the label of public  
11 health pesticides with the statement that it is a violation of  
12 EPA regulations and of the label to state that the pesticide is  
13 safe.

14 Secondly, one resident ran an adverse effects  
15 hotline on the reregistration docket for Prallethrin. They  
16 submitted 14 cases of adverse effects. Given the number of  
17 instances of vomiting, they wonder that if Prallethrin, which is  
18 supposed to cause benign agitation of mosquitos and rapid knock  
19 down, if that may have some human effects such as vomiting.

20 Thirdly, they hit 1150 square miles twice.  
21 Some people think this is the new norm. We wonder if maybe they  
22 should file an Environmental Impact Statement because in the EIS  
23 process, maybe the bees would get some representation. Thank  
24 you.

25 **DR. DANIEL SCHLENK:** Any questions for Mr.  
26 McFadden before he leaves. Would you like to answer any



1 questions? You don't have to. Dr. James?

2 **DR. ROSALIND JAMES:** Could you tell me what  
3 your organization is again.

4 **MR. STEPHEN MCFADDEN:** Independent Scientific  
5 Research Advocates.

6 **DR. ROSALIND JAMES:** Okay, who is that?

7 **MR. STEPHEN MCFADDEN:** Well, it's me, myself  
8 and I, and I also run the website dalcap.org. And I work with  
9 several people in the Dallas Fort Worth area. In 2002 and 2003,  
10 I was living in Dallas, and we got on the city council to try to  
11 put together IPM program for the West Nile virus. So, in 2002  
12 and 2003, I was living there on a daily basis.

13 This year, I cancelled my trip to Dallas when  
14 I saw aerial spraying listed, but I followed basically every  
15 news article on the internet and I work closely with the  
16 activists there.

17 It's just like all of a sudden, government  
18 screwed up, nobody is watching the store and all of the sudden,  
19 they tell the public we are going to be spraying by air in a  
20 couple of days and it's going to hit 1000 square miles.

21 The bees don't like it, we don't like it.  
22 They claim its safe in the all the press and we know that that  
23 isn't supported that the claim of safety fails to consider  
24 individual factors such as genetics, health history, et cetera.  
25 So the term safe is conclusive when you can't make a conclusory  
26 term about 2 million people. Thank you.



1                   **DR. ROSALIND JAMES:** Thank you.

2                   **DR. DANIEL SCHLENK:** Okay. At this point in  
3 time, our time of public commenting is closed. I believe so.  
4 So, before lunch if we could, if we can have the Agency come up  
5 again, and if the Panel has any questions related to the  
6 presentations that were made yesterday, you've had the evening  
7 to sort of mull over those presentations and your question  
8 groups discuss a few of those. If you have any particular  
9 questions to any particular representatives here, now is the  
10 time to do it before we get into our panel question  
11 deliberations. So, Dr. Berenbaum?

12                   **DR. MAY BERENBAUM:** I just have a question,  
13 don't even know to whom it should be directed, but it is for  
14 general information and I apologize if everyone else knows this,  
15 but I don't. Incident reporting - who is, how is that - we've  
16 heard there is state level, federal level, what agency, who has  
17 oversight, how is it regulated?

18                   **DR. THOMAS STEEGER:** The only entity that's  
19 required to report incidents to the Agency is the Regulated  
20 Community Registrants themselves under FIFRA 6A2. The Agency  
21 provides funding to states who are the - we refer to them as the  
22 State Lead Agencies. They have primacy for enforcing labels and  
23 they are responsible for investigating incidents that are  
24 reported to them.

25                   The Agency is aware that many beekeepers feel  
26 that for a variety of reasons incident reports are not making it



1 back to the Agency. The Agency makes clear in its assessments  
2 that the absence if incident reports cannot be construed as the  
3 absence of incidents.

4 So, understanding that there have been  
5 breakdowns in communication and that the number of incidents is  
6 potentially being under reported, there is reluctance by many  
7 beekeepers to report incidents because of fear of offending  
8 growers that they depend on for pollination services and for  
9 honey production. And, there is actually fear of retribution  
10 that bee colonies will be destroyed.

11 That is very unfortunate. But understanding  
12 that there are impediments to reporting incidents, the Agency  
13 has established other means for individuals to contact the  
14 Agency directly through the National Pesticide Information  
15 Center, and they can contact us through beekill@epa.gov. I  
16 monitor the portal for the beekill@epa.gov.

17 We did have the opportunity to record Jim  
18 Doan's bee kill incidents, and we were in contact with the state  
19 of New York through our regional offices to encourage them to  
20 investigate those incidents.

21 **DR. DANIEL SCHLENK:** Any other questions? Dr.  
22 Potter?

23 **DR. THOMAS POTTER:** Yes, I have a question  
24 about the data that was summarized in the document describing  
25 pesticide concentrations in pollen and nectar. And my question  
26 is does that represent the entire set of information that you



1 utilized in the process of establishing a dietary exposure  
2 estimate using T-REX?

3 **MS. KRIS GARBER:** This is Kris Garber. We  
4 used all the available information from the open literature and  
5 unpublished registrant submitted studies that we are aware of.  
6 We did some literature searches, and we searched our database of  
7 submitted studies.

8 We did exclude some studies from the open  
9 literature that didn't provide application rate information  
10 because it was really important for us to be able to link the  
11 mass of the pesticide that was put on the field so that, you  
12 know mass that was observed in the different samples, especially  
13 for the T-REX and for the Briggs' analysis. So, there are some  
14 studies in the literature that we excluded based on application  
15 rate.

16 **DR. THOMAS POTTER:** I got the impression, and  
17 maybe again, that is why I'm following up on this, that there  
18 were registrant submitted studies that contained information on  
19 active ingredient concentrations in pollen and nectar that were  
20 utilized. So, what you're telling me is that is not the case  
21 other than what you describe in the White Paper?

22 **MS. KRIS GARBER:** What I'm saying is that  
23 there were registrant studies that had concentrations in pollen  
24 and nectar, and those are included in the White Paper and they  
25 are summarized in appendices 3 and 4.

26 **DR. THOMAS POTTER:** Right, and again, I will



1 just make a point here that I think there are to me a  
2 surprisingly small number of measurements that are available to  
3 use in making assessments about the dietary exposure here. You  
4 know, I think that is critically important. We will get into  
5 that this afternoon. Maybe I'm jumping the gun here a bit, but  
6 I wanted to confirm what data you had utilized in your  
7 assessments.

8 **DR. DANIEL SCHLENK:** Dr. Pettis?

9 **DR. JEFF PETTIS:** Jeff Pettis. Kind of  
10 following up on that, in looking at the consumption rate of  
11 nectar versus pollen, I just had a feeling that the pollen  
12 consumption rate seemed abnormally low. Can you comment on the  
13 calculations of the average bee consuming nectar versus pollen?  
14

15 **MS. KRIS GARBER:** Well, we - both of the  
16 consumption rates for pollen and nectar were empirically based.  
17 The pollen consumption rates are based on direct counts of  
18 pollen grains in the guts of bees, including adults and larvae.  
19 So, I think those are some pretty confident measures because  
20 they were actually able to, you know, count how many pollen  
21 grains were in each bee.

22 Now the nectar consumption rates are not based  
23 on direct measures of how much a bee eats, either nectar or  
24 honey. So we had to calculate it based on the energetic  
25 requirements of the bees in terms of how much sugar they may  
26 need based on their different tasks. So the nature of the



1 estimate is different.

2 Perhaps that could lead to some differences in  
3 the magnitude of those two. But for a nectar forager that  
4 spends a lot of its time flying, it makes sense that it would  
5 need a lot of sugar to, you know, fulfil its task.

6 **DR. DANIEL SCHLENK:** Dr. James?

7 **DR. ROSALIND JAMES:** This is Rosalind James.  
8 I have a follow-up question to that. Later today we will  
9 probably talk some more about this difference between the pollen  
10 and nectar in the EPA assessment for Tier I. In some ways, it  
11 wouldn't matter because you are essentially saying that the  
12 concentration of pesticide is the same in both, and yet some of  
13 the commenters today were saying pesticide concentration in  
14 pollen is quite a bit higher than in nectar. So, this is an  
15 important question.

16 **MS. KRIS GARBER:** When we were coming up with  
17 the Tier I method for foliar spray applications, that's based on  
18 the tall grass residue of the surrogate for pollen and nectar,  
19 we did actually consider applying some kind of adjustment  
20 factor, which was something that was brought up in the public  
21 comments; but there were some studies where - and I think that  
22 one of them is Choudhary et al., which is summarized --  
23 Choudhary and Sharma I think are the two authors. There were  
24 actually some cases where the concentrations in the pollen and  
25 the nectar were equivalent.

26 So, we thought that you couldn't necessarily



1 assume that the concentrations are always going to be less in  
2 nectar, although it is true that there are several studies where  
3 that is the case. So, that's why we decided to assume that  
4 nectar and pollen were equivalent and that, you know, like Dr.  
5 Potter had pointed out, there isn't a lot of data available for  
6 pollen and nectar. So, we didn't think that we had enough  
7 information to necessarily apply an appropriate adjustment  
8 factor.

9 **DR. DANIEL SCHLENK:** Go ahead.

10 **MR. KEITH SAPPINGTON:** Keith Sappington, EFED.

11 I just want to follow up with one point on those data that they  
12 represent day zero values. So values, you know, measured  
13 immediately after application or within 24 hours, so that is  
14 something to keep in mind with the foliar applications.

15 The other thing is we notice some differences  
16 across plant species. You might think that the availability of  
17 the nectaree to receive the spray droplets would be a factor, so  
18 different plants and different physiologies associated with the  
19 nectarees could come into play. So we looked at this, but we  
20 felt that the range of, basically the variability around the  
21 pollen to nectar ratio was too large to proposed as a method.

22 **DR. DANIEL SCHLENK:** Dr. Fefferman?

23 **DR. NINA FEFFERMAN:** Hi thanks. Along the  
24 same lines, I was actually - I like very much the sort of  
25 energetic analysis of the need for the workers for nectar  
26 consumption and the absence of their being a measurement for



1 that. I was wondering of the similar logic couldn't be applied  
2 to the protein needs of egg-laying queens in order to establish  
3 a rate separate for consumption there.

4 **MS. KRIS GARBER:** I think that's an excellent  
5 point and we could definitely consider that.

6 **DR. DANIEL SCHLENK:** Dr. Pistorius?

7 **MR. JENS PISTORIUS:** I have question. Would  
8 the EPA be in charge and in power to implicate risk mitigation  
9 measures of beyond the action of application of pesticides and  
10 especially, for instance, concerning seed treatment facilities,  
11 prescribing a certain seed quality level? And in addition to  
12 that, for instance like that for non-tilling systems, that there  
13 has to be a herbicide application or removal of flower plants  
14 before application - is that possible? And machine technique as  
15 well, so you could prescribe use of deflectors?

16 **DR. THOMAS STEEGER:** The EPA has been working  
17 with the seed treatment organizations and with the seed  
18 equipment manufacturers as well as the registrants to develop  
19 that very type of mitigation. The label though, is specific to  
20 how the product is applied as opposed to how it might be  
21 distributed in plantars.

22 So, it is very difficult from my understanding  
23 and out my realm for speaking to this issue, but we feel that  
24 the best means of mitigating those potential effects is through  
25 best management practices that would be imposed by industry.

26 **DR. DANIEL SCHLENK:** Dr. Hunt?



1                   **DR. GREG HUNT:** I want to get back to - this  
2 kind of relates to the previous question in a way. I want to  
3 get back to the pollen, the amount of pesticide in pollen,  
4 specifically Clothianidin; the White Paper cites our Krupke et  
5 al. paper as 3.9 parts per billion in the corn pollen that we  
6 analyzed. But we also took pollen from pollen traps on hives  
7 and found up to 88 parts per billion of Clothianidin. And this  
8 was at a time when corn was not being planted.

9                   So, my hypothesis is that this is from talc  
10 dust that had remained on flowers for a while because that kind  
11 of level, we just don't find it in corn pollen that has been  
12 treated, although the bees were bringing in up to 80 percent  
13 corn pollen in the sampling.

14                  **DR. DANIEL SCHLENK:** So, is this a question  
15 for the Agency or a comment?

16                  **DR. GREG HUNT:** I guess this is a comment.

17                  **DR. DANIEL SCHLENK:** Yeah, let's hold the  
18 comments until later. Okay?

19                  **DR. GREG HUNT:** All right, sorry.

20                  **DR. DANIEL SCHLENK:** These are questions the  
21 Agency that will help you in your comments later on. Okay?  
22 That's what this time is for. Any other questions? Okay.  
23 Thanks a lot. Let's break for lunch. We will come back at  
24 12:50, and read in the questions at that time. Thanks.

25                  (WHEREUPON lunch was taken)

26                  **DR. DANIEL SCHLENK:** Question one - and just



1 to save a little bit of time, who's going to be reading the  
2 question in? Who is planning to read the question in? Okay.  
3 You just want to read the letter (a) instead of the whole thing.  
4 I think that part will be sufficient instead of reading the -  
5 so the sub-letters basically throughout I think will be good.  
6 Great, thanks.

7 **MR. THOMAS MORIARTY:** This is Tom Moriarty. I  
8 also understand that you want us to pause after each  
9 subquestion?

10 **DR. DANIEL SCHLENK:** Correct. We will go  
11 subquestion by subquestion.

12 **MR. THOMAS MORIARTY:** Just enough to take a  
13 breath so I can move onto the next one.

14 **DR. DANIEL SCHLENK:** Okay, go ahead.

15 **MR. THOMAS MORIARTY:** Tom Moriarty, U.S. EPA.  
16 Charge question 1, sub (a) - please comment on whether the  
17 assessment endpoints, population size and stability of managed  
18 bees, quantity and quality of hive products, and species  
19 richness and abundance, identified in Table 1, Section 2.2.1 of  
20 the White Paper, are consistent with the Agencies protection  
21 goals. Please include a discussion of any additional assessment  
22 endpoints that may be necessary to meet those protection goals.

23  
24 **DR. DANIEL SCHLENK:** Okay. Our lead  
25 discussant for this particular question is Dr. James.

26 **DR. ROSALIND JAMES:** This is Rosalind James.



1 Since I am answering the first question, I have a couple of  
2 general comments. They won't be long. To the group, I wanted  
3 to commend EPA on the excellent job in writing the White Paper.  
4 It's very thorough and well thought out. It's clear that a lot  
5 of care went into writing this. This proposal is definitely a  
6 step in the right direction. It is very heartening to see EPA  
7 taking on this task with bees.

8 And I have another short comment to the group,  
9 the Panel here. This is a small plea on the use of language,  
10 and in particular, I would like to ask you not to use the words  
11 bees and honey bees synonymously because remember, we are  
12 talking about diversity and we will be talking bees other than  
13 honey bees. So, please say honey bees when you mean honey bees  
14 and when you say bees, mean more than just honey bees. It will  
15 be helpful for me, I know, and probably for EPA as well.

16 With regard to question 1(a), essentially this  
17 question has to do with Table 1 on page 44. So, I am going to  
18 work off of the Table in my answer here.

19 First we are looking at the protection goals  
20 that have been set by EPA and the question is whether or not the  
21 assessment endpoints address the protection goals. So, here I  
22 would like to reiterate about the bees and in question 1 for  
23 provisions of pollination and services. The assessment endpoint  
24 says managed bees. So, we wanted to make sure that EPA is clear  
25 that this means commercial managed bees, which means honey bees,  
26 bumble bees, alfalfa-leaf cutting bees and mason bees and any



1 other bees that may become managed in the future.

2 So when it says population size, we weren't  
3 really sure if this meant colony population size as for honey  
4 bees or actually truly population size, and that really it  
5 should probably be populations in terms of numbers of colonies.

6  
7 Then for production of hive products - the  
8 assessment endpoint is quantity and quality of hive products.  
9 Quantity and quality of hive products is fine endpoint, but  
10 probably quantity would be sufficient, unless quality really  
11 means pesticide-free or low pesticide residues. In this case  
12 quality is an important measure we will see in the next  
13 subquestion.

14 Onto number 3 -- contribution to pollinator  
15 biodiversity. The question really is asking about the  
16 assessment endpoints and whether or not these endpoints address  
17 a protection goal. However, we feel that the protection goal is  
18 very broad. It is sort of like saying we're going to cure  
19 cancer, or we are going to save the world - we are going to stop  
20 global warming. It's just a very, very large goal, and  
21 likewise, the assessment endpoint is very, very broad to species  
22 richness and abundance. Furthermore throughout the White Paper,  
23 there really are no measures for species richness and abundance  
24 taken.

25 So we recommend that the protection goal be  
26 rewritten to read contribution to bee biodiversity. If you are



1 talking about pollinators, this would mean all pollinators,  
2 insect pollinators and mammals and other vertebrates like  
3 humming birds and so on. It just seems beyond the scope of this  
4 White Paper.

5 The honey bees - the entire genus Apis - they  
6 are not native to the Americas, and there is approximately 4000  
7 species of bees that are native to the Americas. New species  
8 are discovered every year. Just in our laboratory alone, we  
9 probably discover two every year. This year, there was a new  
10 species discovered in the middle of New York City. So, that's  
11 how poorly they're known. I mean, there are 4000 species that  
12 are known, but - so, we just wanted you to keep that in mind as  
13 you think about this goal. I think that is the end of what I  
14 have for (a). It is hard for me to separate out (a), (b) and  
15 (c), but we will get to (b) and (c).

16 **DR. DANIEL SCHLENK:** Okay. Our first  
17 associate discussant is Dr. Berenbaum.

18 **DR. MAY BERENBAUM:** I concur.

19 **DR. DANIEL SCHLENK:** And Dr. Pettis.

20 **DR. JEFF PETTIS:** I concur as well.

21 **DR. DANIEL SCHLENK:** Dr. Pistorius?

22 **MR. JENS PISTORIUS:** I concur as well.

23 **DR. DANIEL SCHLENK:** Dr. Ostiguy.

24 **DR. NANCY OSTIGUY:** I concur also.

25 **DR. DANIEL SCHLENK:** All right, so let's open  
26 it up to other Panel members for any other comments for Question



1 1(a). Anything else to add? Okay. Let's - who's going to be -  
2 any questions for clarification for the Agency for that? We're  
3 good? All right, thanks. All right, you want to read in 1(b),  
4 Mr. Moriarty.

5 **MR. THOMAS MORIARTY:** Please comment on  
6 whether the measurement endpoints at the level of the colony,  
7 for examples, colony strength and survival, contamination of  
8 pollen and nectar, and species richness and abundance identified  
9 in Table 1 are consistent with the assessment endpoints  
10 identified in the Table and any additional assessment endpoints  
11 discussed in Part (a) of this question. Please include a  
12 discussion of any additional measurement endpoints that may be  
13 necessary to represent those assessment endpoints.

14 **DR. DANIEL SCHLENK:** And again, Dr. James, you  
15 are our lead discussant for that question.

16 **DR. ROSALIND JAMES:** All right, this is  
17 Rosalind James. So, again we are still working off of Table 1  
18 and now we are on the third column measurement of endpoints and  
19 how these meet the protection goals. So, back again to bee  
20 diversity, 98 percent, probably closer to 99 percent of bee  
21 species are solitary, and thus they do not have colonies. And,  
22 so a measurement of the endpoint colony strength would not  
23 really be measuring these bees directly. We want to make sure  
24 that that's clear. However, if honey bees serve as a surrogate  
25 for all bees, then this endpoint should be fine, but EPA needs  
26 to keep in mind that this is honey bee serving as a surrogate,



1 so it is addressing honey bees and other bees. We also wanted  
2 to add to this colony strength and survival is listed, but we  
3 also wanted to add colony development, which means the growth of  
4 the colony during growth season.

5 For the second item, production of hive and  
6 hive products, again, we recommend removing the word quality and  
7 also we wanted to add other hive products to this list. In  
8 particular, pollen, propolis and royal jelly; so that this  
9 element would now read, "quantity of hive products; and residues  
10 levels in honey, pollen, wax, propolis and royal jelly."

11 And this was partly keeping in mind for honey  
12 bees and then for the native bees as well. Some of these plant  
13 resins and so on are very important to native bees in their  
14 natural habitat. If they are contaminated with pesticides,  
15 especially the larvae would be exposed.

16 So then onto number 3 - contribution to bee  
17 biodiversity. What's currently the measurement endpoints are  
18 colony, strength and survival, and the second element is species  
19 richness and abundance. Colony survival is repeated here from  
20 up above and that's fine, but all my earlier comments about  
21 colony strength and survival apply here as well, and especially  
22 now we are talking about not honey bees, but other bees. So,  
23 those comments are doubly important here.

24 Then species richness and abundance - again,  
25 this is a noble goal, but it's not really included anywhere in  
26 the assessments that occur in the White Paper. There's no way



1 that you can assess species diversity using only one species,  
2 the honey bee. Again, this is a species in addition,  
3 essentially a domestic animal that's not native to any of the  
4 Americas. So, species richness and abundance is - it would be a  
5 good measurement endpoint, but you haven't addressed it at all.  
6 All right, that is the end of my comment.

7 **DR. DANIEL SCHLENK:** Thank you. Dr.  
8 Berenbaum.

9 **DR. MAY BERENBAUM:** I just want to clarify or  
10 emphasize that taking quality out of the measurement endpoint  
11 for hive products does not affect the validity of residue  
12 levels. So, quality independent of residue levels is not really  
13 the issue, but residue levels certainly do affect quality.

14 **DR. DANIEL SCHLENK:** Okay. Dr. Pettis?

15 **DR. JEFF PETTIS:** I concur with what's been  
16 said.

17 **DR. DANIEL SCHLENK:** And Dr. Pistorius?

18 **MR. JENS PISTORIUS:** Same here.

19 **DR. DANIEL SCHLENK:** Dr. Ostiguy?

20 **DR. NANCY OSTIGUY:** I just wanted to add that  
21 in looking at the using of honey bees to measure biodiversity,  
22 one of the things that we discussed are the studies that have  
23 been published on the negative impact of the presence of honey  
24 bees on native species. So, it's a significant consideration  
25 that we not use honey bees as a measurement of biodiversity.

26 **DR. DANIEL SCHLENK:** Okay. Yes, Dr. James?



1                   **DR. ROSALIND JAMES:** This is Rosalind James.

2       I want to add to that. At the foot note of this Table, and I  
3       believe it was said elsewhere in the White Paper also, the very  
4       last sentence says, in addition, protection of the honey bee  
5       would contribute to pollinator diversity indirectly by  
6       preserving the pollination and propagation of many plant species  
7       pollinated by honey bees, which also serve as food sources for  
8       other pollinating insects. Although, we have not been asked to  
9       edit this document, I would recommend deleting that because of  
10      the issue that Nancy brought up that many people - there's  
11      considerable body of research investigating how honey bees  
12      actually compete with native bees. So honey bees in themselves  
13      are not a good measure of bee diversity.

14                  **DR. DANIEL SCHLENK:** Dr. Fefferman?

15                  **DR. NINA FEFFERMAN:** Hi, I'm not a discussant  
16      on this question, so I hope this is still okay. I wanted to  
17      mention that maybe instead of looking just at measurements of  
18      biodiversity also to consider possibly as a measurement, the  
19      genetic variation seen within a species; because long-term  
20      conservation, especially population genetics models tell us that  
21      when we look for effective population sizes, those can be quite  
22      important. So, if we are looking at end measures of quality in  
23      the long term instead of the short term, along the same lines of  
24      biodiversity, genetic variation might be useful.

25                  **DR. DANIEL SCHLENK:** Thank you. Any other  
26      comments from panel members? Okay. Dr. James, I am sure that



1 you are going to get Dr. Fefferman's comments into that? Okay.  
2 Great. All right. Let's go to the Agency. Any questions of  
3 clarification? Okay. Great. Moving right along. Okay. Mr.  
4 Moriarty, you want to read in 1(c) please?

5 **MR. THOMAS MORIARTY:** Thank you. Question 1,  
6 subpart (c) - please comment on whether the measurement  
7 endpoints at the level of the individual bee, individual adult  
8 and larval survival, queen fecundity, brood emergence success  
9 and worker longevity, identified in Table 1 are consistent with  
10 the assessment endpoint identified in the Table and any  
11 additional assessment endpoints discussed in part (a) of this  
12 question. Please include a discussion of any additional  
13 measurement endpoints that may be necessary to represent those  
14 assessment endpoints.

15 **DR. DANIEL SCHLENK:** And Dr. James, lead  
16 discussant.

17 **DR. ROSALIND JAMES:** All right, so we are  
18 still on Table 1 and now looking at the fourth column  
19 measurement endpoints at the individual level and how this meets  
20 the protection goals. The items listed as measurement endpoints  
21 are very similar among the three different goals, and therefore  
22 I am going to discuss them all together.

23 Individual worker survival is a very good  
24 measure. Larval survival should also be included. It's not  
25 listed here. Larval bioassays, I think, are going to be a very,  
26 very important contribution to understanding the possible



1 effects of pesticides on bees.

2 We suggest here using the word larvae and not  
3 brood, so that it's clearly inclusive of more than just honey  
4 bees. Brood is a word that is used basically for different  
5 larval stages and pupae of honey bees, even though the test will  
6 be for honey bees.

7 We also recommend adding larval development  
8 time as one of your measurement endpoints; in particular, the  
9 time from egg lay to when a cell is capped. The negative  
10 effects of some pesticides such as insect growth and regulators  
11 may be able to be best detected using this sublethal measure.

12 Also, we were not clear what is meant by brood  
13 success, which is on the products of the hive, so it's on the  
14 second box. We recommend changing this to larval survival and  
15 delayed development instead of brood success, which is  
16 essentially a repeat of what I just said above for the previous  
17 box.

18 Brood size is a good measure, and there are  
19 fairly standard ways for measuring this in a hive. However,  
20 some people might think that this means individual larval size.  
21 So we recommend changing the wording to brood nest size and  
22 define it as the area of the sealed brood in a colony.

23 Queen fecundity is a very useful endpoint, but  
24 at this time, it's difficult to measure. Perhaps other people  
25 on the Panel would want to make some comments on fecundity and  
26 the utility of using this and how it might be measured. That's



1 the end of my comments.

2 **DR. DANIEL SCHLENK:** Dr. Berenbaum?

3 **DR. MAY BERENBAUM:** One of the parameters that  
4 are difficult to quantify is worker bee longevity because it  
5 tends to vary over the life cycle of the bee. The assays that  
6 exist remove the bee from its social context, so their meaning  
7 of the results is difficult to ascertain. So, in terms of  
8 reliable endpoints, worker bee longevity may present some  
9 experimental challenges.

10 **DR. DANIEL SCHLENK:** Okay. Dr. Pettis you're  
11 next.

12 **DR. JEFF PETTIS:** Just a couple of comments.  
13 One is for protection goal three and the measurement endpoints.  
14 Since it is a biodiversity or bee biodiversity, we are  
15 recommending that it be bee biodiversity protection goal. Then  
16 brood success for solitary pollinators would be a very excellent  
17 measure if you were testing solitary bees; the female's brood  
18 production would be an excellent measure and endpoint.

19 Then last, the queen fecundity issue, I see  
20 that as a research gap, that we don't have adequate ways to  
21 measure queen fecundity at this time.

22 **DR. DANIEL SCHLENK:** Thank you. Dr.  
23 Pistorius?

24 **MR. JENS PISTORIUS:** Adding to the queen  
25 fecundity, I think we have a possibility of an indirect  
26 measurement by the assessment of brood and colony development.



1 That is the only method available.

2 **DR. DANIEL SCHLENK:** Okay. And Dr. Ostiguy?

3 **DR. NANCY OSTIGUY:** I concur with the  
4 comments.

5 **DR. DANIEL SCHLENK:** Okay. Any other panel  
6 members want to chime in on this one? Yes, Dr. Hunt.

7 **DR. GREG HUNT:** I am just responding to what  
8 May Berenbaum said. Worker longevity does present some  
9 problems, but if you could measure it as kind of an average  
10 worker longevity if you, for example, mark the bees and then  
11 take photographs of the comb, so that could be a possibility.

12 **DR. DANIEL SCHLENK:** Dr. Berenbaum?

13 **DR. MAY BERENBAUM:** Yeah, just to stay it will  
14 present experimental challenges. That value is still going to  
15 vary depending on season and timing and all kinds of other  
16 factors. I'm not saying not to use it, but it can be hard to  
17 standardize.

18 **DR. DANIEL SCHLENK:** Yes, Dr. Tarpy?

19 **DR. DAVID TARPY:** This is Dave Tarpy. I would  
20 probably feel a little more comfortable rather than using kind  
21 of a more restrictive term of queen fecundity in this context to  
22 really mean something more like queen reproductive potential,  
23 which incorporates other things. Fecundity, at least to me - I  
24 could be a minority of one here - but fecundity is really kind  
25 of egg laying rate. I think there are other factors dealing  
26 with queen reproductive success and reproductive potential that



1 could be important and captured here.

2 **DR. DANIEL SCHLENK:** I see a lot of heads  
3 nodding, so I assume that's a consensus sort of statement there.  
4 Okay. Any other comments on 1 (c)?

5 Okay. Hearing none let me go back to the  
6 Agency. Do you have what you need for that particular question?  
7 Fabulous. Let me just say thanks to Dr. James and that group  
8 for getting together and sort of expediting things. I really  
9 appreciate that. Okay. Move on to charge question 2. Again,  
10 if you wouldn't mind reading the subletters for 2, that would be  
11 great.

12 **MR. KEITH SAPPINGTON:** Keith Sappington, EPA.  
13 Charge question 2, subpart (a) - please comment on whether the  
14 conceptual models depicted in Figures 4 through 8 are consistent  
15 with the protection goals and assessment endpoints identified in  
16 Table 1 and as discussed in question 1.

17 **DR. DANIEL SCHLENK:** Okay. And lead  
18 discussant for this particular question is Dr. Ostiguy.

19 **DR. NANCY OSTIGUY:** First comment - the  
20 conceptual models are very good. Lots of detail. We have  
21 comments that will help, I hope, fine tune things. I am going  
22 to start with some general comments, and then I have specific  
23 comments on specific figures.

24 At least according to our reading at this  
25 point, none of the models include wax contamination, most  
26 specifically for larvae. They are sitting in contact with wax



1 during the larval stage. Wax is included, sort of after the  
2 hive worker, the nurse bee, but it needs to also be looked at as  
3 an exposure media for the immatures.

4 One of the things that came up yesterday was  
5 the comment about adding the RQs for oral and contact for the  
6 larva. And because of, you know, the specific lifestyle of the  
7 larva, that probably would be a good idea to combine that  
8 information rather than working just from the oral contact.

9 One of the other comments, again dealing with  
10 the larval stage, most of our comments - a lot of the comments  
11 are on larva because we are still, of course, developing the  
12 proper protocols for evaluating them -- is to actually look at  
13 the total consumption of food during the larval stage rather  
14 than a per day consumption. It would then give you a subacute  
15 test, but a more realistic test since the quantities of food  
16 consumed over time change rather significantly and the types of  
17 food change. So, one value might actually be more useful.

18 This is probably going to come up in a number  
19 of locations. Since I'm talking about larva, what will have to  
20 be chosen is a particular - well, it probably would be the day  
21 that capping occurs, is that you would actually use the weight  
22 rather than a larval measurement. So, a lot of the things that  
23 we have talked about are done as a quantity of exposure per bee.

24  
25 While this has been the way that the  
26 apiculture research community has reported exposures in the



1 past, it's not really appropriate when we are going to be using  
2 honey bees as a surrogate because the other non-honey bees are  
3 certainly different in sizes.

4 A more general comment, the feeling is that  
5 the assorted dotted lines should be changed to be solid lines.  
6 They should not be removed at this point as less important  
7 exposures or the receptors, the receiving end.

8 One of the reasons for wanting to include more  
9 exposure media, more receptors, is that we are using honey bees  
10 as a surrogate and there are going to be exposures that are  
11 potentially important to non-honey bees that otherwise would not  
12 be captured. So for instance, leaf cutter bees - please correct  
13 me if I get this wrong - leaf cutter bees used for alfalfa  
14 pollination, at this point, we probably bring in as many or  
15 nearly as many leaf cutting bees to pollinate alfalfa, some  
16 coming from as far away as Canada.

17 These bees are in contact with soil. So  
18 excluding soil when we are doing the evaluation is not a good  
19 idea. In addition, since we are going to be using honey bees as  
20 a surrogate for, let's say tube nesting, solitary bees, they use  
21 mud for their partitions. So, we should really include that  
22 soil exposure when we are looking at the honey bees. Generally,  
23 the group did agree that, you know, the matrices that you  
24 created are appropriate for both honey bees and other assorted  
25 native bees.

26 Two specific comments - figure 6 - there



1 should be a line going from drift abraded seed coating as a  
2 source to the exposure media residues in or on pollen, nectar,  
3 exudates and honey dew. Then figure 7 needs to include dust as  
4 an exposure media. Then, residues - it should read for the  
5 exposure media of pollen, nectar and exudates, it should be  
6 residues in or on since we are looking at soil as a possible, or  
7 dust as a media. Those are the comments that I have at this  
8 point.

9 **DR. DANIEL SCHLENK:** Thank you. Dr. Hunt, you  
10 are our first associated discussant.

11 **DR. GREG HUNT:** I concur.

12 **DR. DANIEL SCHLENK:** Thanks. Dr. Pistorius?

13 **MR. JENS PISTORIUS:** I concur, except one  
14 comment for figure 6. As you have said, there's one line from  
15 drift abraded seed coating to residues in pollen and nectar, we  
16 are talking about residues on planted seeds, but what we mean is  
17 on off crop lands.

18 **DR. DANIEL SCHLENK:** Okay.

19 **MR. JENS PISTORIUS:** Or in crop lands if there  
20 is no tillage.

21 **DR. DANIEL SCHLENK:** Okay. Yes, Dr. Ostiguy?

22  
23 **DR. NANCY OSTIGUY:** Nancy Ostiguy. I did not  
24 understand and didn't hear what you wanted in there.

25 **MR. JENS PISTORIUS:** Okay. With this line  
26 drift of abraded seed coating, as you said, we want the line



1 drift to residues in pollen, nectar and exudates and honey dew;  
2 but the box above is talking about residues on planted seeds.  
3 But what we mean is not drift on the planted seeds nor the  
4 secreted seeds that we bring into the soil, but we want to look  
5 at residues in pollen and nectar off crop or if there are  
6 flowering weeds in the area which is sown. Does that clarify?

7 **DR. DANIEL SCHLENK:** Do you have that  
8 information? Perhaps just a written component of your comments,  
9 if you could send that, that would probably be appropriate as  
10 well just to confirm. Okay. Dr. Potter?

11 **DR. THOMAS POTTER:** I concur with everything.  
12 I just wanted to add one small detail, and this is in regard to  
13 the soil exposure. I believe that one thing we may want to  
14 consider is the treated soil as a source of dust via wind  
15 erosion and eroded particles from soil via wind can be strongly  
16 enriched if pesticides and active ingredients are implied to  
17 them. So, that might be an added twist in terms of the exposure  
18 issues that we may be addressing in the case of pesticides and  
19 soil.

20 **DR. DANIEL SCHLENK:** Okay. Thanks. And Dr.  
21 Schwab?

22 **DR. PAUL SCHWAB:** Yes, I only have one comment  
23 and that's on figure 6. With the seed based translocation, the  
24 box from the residue on planted seeds, this is -- just that term  
25 seed based translocations is a bit confusing to me in itself.  
26 I'm not really sure what that means, and it could be - I don't



1 know if this means that the seed itself is somehow assimilating  
2 the pesticide directly from the dust or the coating I mean, but  
3 I think it would help to clarify if we could consider the seed  
4 coating as becoming part of the soil as well, at least some  
5 fraction of that, and then being subject to uptake through the  
6 roots. Maybe that's what is implied already, but it wasn't  
7 clear to me. But otherwise, I concur with all the other  
8 statements.

9 **DR. DANIEL SCHLENK:** Okay. Other Panel  
10 members? Yes, Dr. Berenbaum.

11 **DR. MAY BERENBAUM:** Did the group discuss  
12 whether there is a significant possibility of exposure via  
13 contamination of resins used for propolis, among other things,  
14 populus as a biofuel crop tree and is a common source for bees  
15 to harvest propolis materials?

16 **DR. DANIEL SCHLENK:** It would appear not.  
17 Okay. Dr. James and then Dr. Hunt.

18 **DR. ROSALIND JAMES:** I've a few different  
19 comments - it's a little bit hard to separate out the different  
20 questions. I will be addressing some of this in the next  
21 question. I am probably the proponent here for the non-Apis  
22 bees. These diagrams are a good place where the non-Apis bees  
23 don't show up. Dr. Ostiguy already talked a little bit. I just  
24 wanted to clarify some of the comments she made.

25 So, the leaf-cutter bees will be using leaf  
26 pieces and the larvae will be growing up in an environment with



1 leaf pieces. So residues that are on these leaf pieces would  
2 have a different experience for alfalfa-leaf cutting bees than  
3 for honey bees. Honey bees would be that foraging adults would  
4 land on them as opposed to larvae being raised inside them. The  
5 majority of solitary bees are actually ground-nesting bees.

6 So, we heard today with public comments and in  
7 some of the public comments, and again it shows up over and over  
8 in the diagrams that bees do not come into contact with soil.  
9 Well, the majority of the wild bees grow up in the soil.

10 And there are lots of models, some of which  
11 are presented here about tracking what the soil residues might  
12 be and those could be applied, perhaps, to include in-contact  
13 toxicity test with larvae. So again, in-contact toxicity tests  
14 on larvae are often excluded because honey bee larvae would not  
15 be exposed to the soil, but many of the solitary bee larvae  
16 would.

17 Also, I would like to reiterate a comment that  
18 was made yesterday in one of the questioning that what about  
19 nonsystemic soil applied pesticide. For the same reason the  
20 bees would actually be exposed to the soil, so even though it is  
21 not a systemic pesticide, it could have some affect on the  
22 non-Apis bees.

23 Throughout the discussions yesterday, I was  
24 kind of struck by a very crop-centric view, but we are actually  
25 talking about an environmental impact and we have flowering  
26 plants in the crops that are not part of the crop. You can have



1       overspray and so on, and the bees would be visiting some of  
2       these.

3                       I am not exactly sure where to put it into  
4       these diagrams, but certainly flowering weeds, or in my  
5       laboratory - I work for the Agricultural Research Service, and I  
6       know some of the other folks here are really trying to encourage  
7       farmers to intentionally plant flowering plants to increase the  
8       bloom period and the blood resources for bees. How do we  
9       include that in a pesticide assessment if there is overspray of  
10      pesticide?

11                     Pesticides are a very big problem for us in  
12      trying to introduce non-Apis bees into farming crop systems.  
13      These are some of the areas. That's it.

14                     **DR. DANIEL SCHLENK:** Thanks. Dr. Hunt?

15                     **DR. GREG HUNT:** I don't know if -- this  
16      comment may not be as important, but in talking about what May  
17      Berenbaum was saying about propolis, this is collected from  
18      typically trees that are not directly exposed to pesticide, but  
19      propolis in the hive could absorb pesticides and residues could  
20      be there.

21                     **DR. DANIEL SCHLENK:** Great. Dr. Ostiguy, I  
22      see you typing this up, so I'm assuming you are putting all this  
23      in. Great. Any other Panel members? Dr. Tarpy?

24                     **DR. DAVID TARPY:** This is Dave Tarpy. I've  
25      noticed on these flowcharts, which I agree are very good and  
26      they also are very kind of Apis centric and don't quite capture



1 some of the subtleties of the solitary bees. But with that  
2 said, the main receptors in these flowcharts as they ought to be  
3 are the foraging bees. In doing so, the arrows that point to  
4 them really, it's implying that the vast majority of the  
5 ingestion is with the foraging bees.

6 I know that the presentations really focus on  
7 that with nectar and is true, but really they are transporting,  
8 but not always ingesting, especially when it comes to pollen.  
9 So, there are some subtleties where hive bees, for example --  
10 the nurses are kind of the direct consumers, the ones that  
11 ingest pollen, for example, rather than the nurse bees. So, I  
12 don't know if that really affects the overall concept of these,  
13 but there is a division of labor when it comes to ingestion. It  
14 doesn't all go through the foragers per se.

15 **DR. DANIEL SCHLENK:** Okay. Thank you. Any  
16 other comments from the Panel members? Okay. Let me go to the  
17 Agency - any questions or clarification?

18 **MR. KEITH SAPPINGTON:** Keith Sappington. I  
19 just have clarifying question on the figures. Do you want us to  
20 move the wax and propolis, which are found in the figures to a  
21 different location?

22 **DR. DANIEL SCHLENK:** Who wants to address  
23 that? Dr. Ostiguy, do you want to try to address that?

24 **DR. NANCY OSTIGUY:** Yes, I will try - this is  
25 Nancy Ostiguy. Yes, because it is an exposure media for larvae  
26 whereas where it is right now is a receptor, at least the way



1 that I'm reading it, wax seems to be incidental?

2 **MR. KEITH SAPPINGTON:** Keith Sappington. Yes,  
3 in terms, I think I understand the comment now that along the  
4 boxes that are listed as receptors, we have receptors, but we  
5 also have essentially exposure media that are relevant to those  
6 receptors, and wax and propolis is listed as an exposure media  
7 for the brood, for the larvae. But that is not necessarily  
8 tracking with the bold - is that the question that that's not  
9 tracking with where the exposure media are listed above it?

10 **DR. DANIEL SCHLENK:** Dr. James?

11 **DR. ROSALIND JAMES:** Yeah, I would like to  
12 address that. I agree with what you suggested actually that  
13 moving the honey wax and propolis as an exposure media instead  
14 of being an attribute change - I mean, it may also be an  
15 attribute change, but that's more with human. Here? I think he  
16 meant here. Oh, we don't know what you mean. Is it here or  
17 here? At receptors or attribute change? You have it in two  
18 places.

19 **DR. NANCY OSTIGUY:** This is Nancy Ostiguy. I  
20 think that we were talking about putting wax and propolis as an  
21 exposure media separate from and maybe in addition to where you  
22 have it right now in the receptors.

23 **MR. KEITH SAPPINGTON:** Okay. I can  
24 understand. Thank you.

25 **DR. DANIEL SCHLENK:** Okay. Everybody clear on  
26 that? Oh, Dr. James?



1                   **DR. ROSALIND JAMES:** I'm the slow one, in the  
2 head here, I guess. To me, you don't really have it as a  
3 receptor. You have it on top of the arrow, what does that mean?  
4

5                   **MR. KEITH SAPPINGTON:** Yes, on top of the  
6 arrow means a mechanism by which, in this case it would be the  
7 bee brood and the queen would be exposed, so they would be  
8 exposed via brood provisions, contact with wax, propolis and  
9 royal jelly.

10                   So, it was just to rather than have a blank  
11 arrow going to them from the hive bees, nurse, worker, drones,  
12 which all participate in processing that material, it was just  
13 to illustrate how they would be exposed. But it could easily be  
14 put above under exposure media and avoid that confusion.

15                   **DR. ROSALIND JAMES:** This is Rosalind James.  
16 It could be in both. I mean, because it really is then an  
17 exposure media even by your explanation and by putting it on the  
18 arrow, it just sort of explains how it is an exposure media.

19                   **DR. DANIEL SCHLENK:** Dr. Ostiguy?

20                   **DR. NANCY OSTIGUY:** This is Nancy Ostiguy.  
21 One of the reasons in addition to placing wax and propolis as an  
22 exposure media is that those particular materials are recycled  
23 by the workers. So, they're going to then repeatedly be taking  
24 it up and placing it back down again.

25                   **DR. DANIEL SCHLENK:** Dr. Hunt?

26                   **DR. GREG HUNT:** I hate to complicate things,



1 but in the exposure media could also have an arrow down to the  
2 hive bees, all the adults, because they are exposed by the wax.  
3 Their exoskeleton dissolves liquids.

4 **DR. DANIEL SCHLENK:** Yes, Dr. Ostiguy? And  
5 then Dr. Berenbaum?

6 **DR. MAY BERENBAUM:** And to complicate things  
7 further; the royal jelly arrow goes from hive bees to queens,  
8 but in fact larvae eat royal jelly too. So, all three of those,  
9 brood provisions, wax propolis, royal jelly, should go to that  
10 joint line because they go to both queens and to regular worker  
11 grubs.

12 **DR. DANIEL SCHLENK:** Dr. Ostiguy?

13 **DR. NANCY OSTIGUY:** This is Nancy Ostiguy. I  
14 think we've talked a little bit about that yesterday where there  
15 was sort of an - because the conceptual model just would get  
16 incredibly messy if we had lines all over the place, which might  
17 be the argue for having provisions, wax, propolis, royal jelly  
18 up in the exposure media maybe to clarify that everything is  
19 moving.

20 **DR. DANIEL SCHLENK:** Okay. Dr. James?

21 **DR. ROSALIND JAMES:** I'm hoping to simplify,  
22 not complicate here. So, maybe your bee brood and your queen  
23 boxes could be one box called larvae, queen and worker, drone  
24 larvae, all of them.

25 **DR. DANIEL SCHLENK:** Okay. Let me go back to  
26 Keith. It sounds like you have a few options here.



1                   **MR. KEITH SAPPINGTON:** I think we understand  
2 some of the confusion. Should the Panel have time or feel it  
3 useful to actually suggest the modifications in the report or  
4 have a revised figure in the report that might also clarify  
5 things. And we could even provide the electronic versions if  
6 necessary because it takes time to.

7                   **DR. NANCY OSTIGUY:** This is Nancy Ostiguy.  
8 That would be wonderful, and then we can make sure that we are  
9 clear on what we are trying to tell you.

10                  **DR. DANIEL SCHLENK:** If you would email that  
11 to Fred that would be great and then he can distribute that.  
12 Other than that, everything is clear? Clear as honey? Sorry.  
13 I was going to say mud, but - all right.

14                  We are ready to move onto question three. Who  
15 is going to be reading that in? Okay. Mr. Moriarty? Again,  
16 please read (a) if you wouldn't mind.

17                  **MR. THOMAS MORIARTY:** Question 3, subpart (a)  
18 - please comment on the extent to which the assessment of risk  
19 to the honey bee may serve to meet the protection goals  
20 identified in the White Paper, that is protection of pollination  
21 services, protection of honey and hive product production, and  
22 protection of pollinator diversity.

23                  **DR. DANIEL SCHLENK:** Okay. Dr. James, you are  
24 the discussant.

25                  **DR. ROSALIND JAMES:** This is Rosalind James.  
26 I want to make a request. I want to address (a) and (b)



1 together. They are hard for me to separate. Can I do that?

2 **DR. DANIEL SCHLENK:** Sure.

3 **DR. ROSALIND JAMES:** Do you want to read (b)  
4 also?

5 **Dr. DANIEL SCHLENK:** Do you want to read (b)  
6 in also then if we are going to do that?

7 **MR. THOMAS MORIARTY:** 3(b) - until guidelines  
8 are developed for testing non-Apis species of bees, please  
9 comment on the extent to which the honey bee may or may not  
10 serve as a reasonable surrogate for non-Apis bees. Given the  
11 differences in lift history, strategies, and potential different  
12 sensitivities to pesticide toxicants, please include a  
13 discussion of which types of non-Apis bees may be particularly  
14 well represented by either the individual level or the colony  
15 level endpoints identified in Table 1 of the White Paper, as  
16 well as which types of non-Apis bees may not be well represented  
17 and therefore may be the focus of potential research areas in  
18 the future.

19 **DR. DANIEL SCHLENK:** Okay. Dr. James?

20 **DR. ROSALIND JAMES:** This is Dr. James. To me  
21 these two questions kind of go together. Due to the large  
22 diversity of non-Apis bees, I understand the EPA cannot test a  
23 significant number of them and furthermore, most of them are  
24 wild and they are not in production in any way and this makes it  
25 difficult to do Tier I types of testing.

26 So, I think that honey bees probably could



1 make a good surrogate species to help represent the  
2 susceptibility of other bees, but I would implore EPA to  
3 recognize that it is a surrogate species that represents other  
4 bees. This then affects how you - this part addresses (a), in  
5 which how you do the risk assessment. So, when you come up with  
6 your flow diagrams and when you come up with your measures of  
7 exposure and so on, you have to keep these other bees in mind as  
8 well as a honey bee.

9 The biology of these other bees is diverse and  
10 great and it's hard to know about all of the possibilities, but  
11 I think there are some basic principles that you could learn.  
12 We discussed some of them already, exposure to leaf pieces of  
13 the larvae and nesting in the ground. Another one is the  
14 seasons in which they fly are going to be very different than  
15 for honey bees.

16 But there are some things that also have to be  
17 kept in mind where honey bees may not be very good surrogates  
18 for representing these other bees. So, as a stop gap measure,  
19 maybe honey bees are adequate, but there has been discussion  
20 within the group of whether really you ought to include some  
21 other solitary bee to help keep in mind some of the particulars  
22 that occur with solitary bees.

23 In particular, we discussed that honey bees,  
24 because of their colony behavior and the fact that the colony  
25 lives for multiple years, they store food and they have large  
26 number of workers and they are able to change status of the



1 workers from being nurse bees to foragers and so on, to  
2 compensate for various environmental factors. So, they can ride  
3 through essentially bad times or they can withstand a certain  
4 amount of mortality among the workers.

5 But with solitary bees or something like most  
6 of the social non-Apis bees that have very small colonies or  
7 their colonies are annual, they don't have the ability to  
8 withstand these kinds of stresses. An example would be with  
9 pollen storage.

10 With your solitary bees and with bumble bees  
11 too, the bees go out and collect pollen - I will back up. With  
12 solitary bees, the female provisions each one of her larvae. So  
13 the female is going to go out and she is going to have one whole  
14 provision made for that larva at a particular time. There may  
15 be a pesticide exposure out of that time. The entire life of  
16 that larva is going to be exposed to that pesticide. A later  
17 larva may not have any exposure to pesticide if it's a  
18 short-lived pesticide out in the field.

19 Whereas with honey bees, maybe they could  
20 avoid the pesticide and live on reserves within the colony if  
21 there is an avoidance mechanism - so their exposure would be not  
22 as forced as it is with the solitary bees.

23 We also had a considerable amount of  
24 discussion about the pollen and how much pollen the honey bees  
25 consume. I think over lunch time, we resolved that. Your  
26 numbers are probably not that inaccurate, they are probably a



1 good estimate. What threw us off was your use of nectar instead  
2 of honey. If we take your nectar numbers and divide them by  
3 three, then we come up with a more reasonable estimate of honey  
4 consumption.

5 With your solitary bees and your bumble bees,  
6 to some extent they don't consume as much honey. They don't  
7 consume any honey - bumble bees make some honey. It's not as  
8 processed as honey bee honey. Your solitary bees generally  
9 don't have honey. They just collect nectar and the primary food  
10 source is really going to be pollen. So, probably more pollen  
11 based.

12 When I look at numbers from my laboratory, I  
13 have numbers from this summer and I know what size my pollen  
14 provisions are. I divide them by 14 days, that's how long the  
15 larvae take to live. I come up with pollen consumption rates  
16 that are similar to what you're finding for honey bees. These  
17 bees are much smaller, but they are eating about the same amount  
18 of pollen per day.

19 So, another bee such as the *Osmia lignaria*,  
20 the blue orchard bee, which has a size more comparable to honey  
21 bees, are probably going to eat more pollen than honey bees.  
22 So, there is probably more pollen consumption in the non-*Apis*  
23 bees; and certainly the ratio of pollen to nectar is going to  
24 probably be higher than it is with honey bees.

25 Again - I don't want to reiterate too much,  
26 but I'm kind of bent out of shape a little bit about not doing



1 contact toxicity to solar soil-applied pesticides - I think that  
2 should be included.

3 For the honey bee test, even though honey bees  
4 will never be exposed probably to soil pesticides, other bees  
5 are likely to be exposed to soil pesticides. So, there should  
6 be some contact toxicity done for those.

7 The other big difference is some of your  
8 solitary bees are going to be more specialists than honey bees.  
9 The honey bees are what we call generalist bee. They are  
10 probably the extreme of generalist bees. Bumble bees also are  
11 fairly general. They will forage on most any kind of flowering  
12 plant. Not any kind of flowering plant, but they have a very  
13 broad host range. Your solitary bees tend to have a narrower  
14 host range.

15 So with the mason bees, the *Osmia lignaria*,  
16 they tend to forage mainly on fruit trees or flowering trees.  
17 They are not going after only one plant, but there's a much  
18 smaller group of plants that they go after. This will be true  
19 for many of your native bees. So, they are going to be very  
20 specialized. Somehow, this should be taken into consideration  
21 in the risk - what do you call -- the risk management aspect I  
22 think, if I understand the difference between risk assessment  
23 and risk management.

24 So when you are depending on the pesticide and  
25 where it's being applied, this may affect some bees more than  
26 others. So, perhaps the honey bee could be the surrogate in



1 your Tier I bioassay, but in your risk assessment you need to  
2 include what specialist bees might be exposed under a particular  
3 circumstance. Especially - I mean not all applications are  
4 agricultural. So, we spray for mosquitos, we spray for forest  
5 pests and so on.

6 Again, if you were to include one other bee, I  
7 thought about this question. Probably the easiest bee to  
8 include in addition to honey bee, and I don't think it's just a  
9 personal bias on my part, but would be Megachile rotundata,  
10 which is the alfalfa-leaf cutting bee. You can buy it by the  
11 gallon.

12 Doing bioassays with them really is no more  
13 difficult than honey bees and probably easier, but they aren't  
14 honey bees, so you can't treat them exactly the same as honey  
15 bees. Certainly adult bioassays would be easy because you can  
16 buy the bees by the gallon, incubate them and emerge them as  
17 adults.

18 Doing larval tests will be more difficult  
19 because like honey bee, you will have to collect larvae out in  
20 the field and that can only be done during the time that the  
21 bees are pollinating alfalfa. So, that's more difficult, but  
22 you could include adult alfalfa-leaf cutting bee or even blue  
23 orchard bee. You can buy those, but they are a little more  
24 expensive and they are not by the gallon, they are by the bee.  
25 So, they are about 50 cents a bee.

26 At least - you know one thing, keep that in



1 mind I guess as you go through the assessment. Something to  
2 force you to keep in mind, not the particularly of only honey  
3 bees if you want to use honey bees as a surrogate.

4 **DR. DANIEL SCHLENK:** Okay. Thanks Dr. James.  
5 Dr. Berenbaum?

6 **DR. MAY BERENBAUM:** I concur.

7 **DR. DANIEL SCHLENK:** And Dr. Pettis?

8 **DR. JEFF PETTIS:** I just have to add a bit to  
9 the using honey bees as a surrogate. It is somewhere along the  
10 lines of Dr. James's comment. Just that, I think two genera,  
11 Osmia and Megachile, both would make excellent things for Tier I  
12 testing. They can be used; you can do direct toxicity testing  
13 on adults and get some idea about the degree to which honey bees  
14 can serve because protection goal 3 is about bee biodiversity.  
15 So really to get at that, some testing using at least one or  
16 both of those species might be valid.

17 **DR. DANIEL SCHLENK:** And Mr. Pistorius.

18 **MR. JENS PISTORIUS:** And I would like to add  
19 those are probably also the easiest species to go to a higher  
20 tier tests, you want semi-field test, you can also use them.

21 **DR. DANIEL SCHLENK:** Okay. Other Panel  
22 members? Dr. Ostiguy?

23 **DR. NANCY OSTIGUY:** This is Nancy Ostiguy. I  
24 would like to add that we are sort of tossing out non-honey bee  
25 species that are useful for testing. Bumble bees are actually  
26 quite good for Tier II, because you can actually confine them



1 and they forage normally.

2 **DR. DANIEL SCHLENK:** Mr. Pistorius?

3 **MR. JENS PISTORIUS:** Except one exception, the  
4 brood development is very difficult to assess because there are  
5 multiple layers of brood and you have no way of assessing them.  
6 That is why it is easier with Megachile or Osmia.

7 **DR. DANIEL SCHLENK:** Yes, Dr. James?

8 **DR. ROSALIND JAMES:** Why would brood  
9 assessment be so difficult? I mean you can put them in small  
10 colonies that you can open up and look at them.

11 **MR. JENS PISTORIUS:** Because you cannot  
12 estimate the exact number of larvae and you cannot watch the  
13 individual development of larvae. When they make all those  
14 piles, one across the other, you don't know what is happening.

15 **DR. DANIEL SCHLENK:** That was Mr. Pistorius.  
16 Okay. Any other Panel input here? Okay. Let me go to the  
17 Agency in terms of questions or clarification. Got a lot of  
18 comments here. Okay. So that wraps up charge question 3.

19 Moving right along. Let's go to charge  
20 question 4. If you could read letter (a) there would be great.

21 **MS. KRIS GARBER:** This is Kris Garber. Charge  
22 question 4, subpart (a) - please comment on the strengths and  
23 limitations of the proposed approach for assessing contact  
24 exposures to honey bees in Tier I exposure assessments, i.e. 2.7  
25 micrograms a.i. per bee per 1 pound a.i. per acre, which is  
26 based on the honey bee specific maximum concentration reported



1 by Koch and Weisser, 1997.

2 **DR. DANIEL SCHLENK:** Okay. Our lead  
3 discussant for that is Dr. Schwab.

4 **DR. PAUL SCHWAB:** Okay. Thank you. This is  
5 Paul Schwab. This is a very important consideration because  
6 foliar sprays can result in some of the very highest levels of  
7 residues other than the foliar spray diet, and thus the highest  
8 level of exposure to the foraging bees.

9 Well, there is a lack of data for bee contact  
10 and general exposure to key pesticides, so some sort of  
11 surrogate is necessary. That was the motivation, I'm assuming,  
12 behind this study in the first place.

13 The approach that's being used has a lot of  
14 merit for a lot of reasons, but it also some limitations.  
15 Clearly in the absence of any data at all, and we will get to  
16 this in part (b), then a model would be necessary, and we will  
17 talk about that in a moment.

18 Some of the strengths of this approach would  
19 be that the surrogate tracer was not being used anywhere else in  
20 the region, so there would be - there were a lot of concerns in  
21 other studies about bees going outside the region of control and  
22 getting into pesticides that weren't part of the study.

23 That wouldn't be the case with this particular  
24 tracer because it's unique. This would not be applied to crops  
25 under any circumstances. And so, these compounding factors are  
26 eliminated. The concentration reported was the highest value



1 that was normalized and that should be fairly protective.

2 Some of the weaknesses would be that it is a  
3 bit of an old study - a 15-year-old study. It doesn't mean that  
4 it has no value, but it's possible that some updating could be  
5 in order.

6 The compound that was used was a sodium  
7 fluorescein. It's a fluorescent dye. It's not really one of  
8 the pesticides we look at obviously. It may not even have many  
9 of the same properties that these pesticides would have, but  
10 nevertheless, it probably is a reasonable start for a surrogate  
11 and under these circumstances.

12 Bees were actually foraging during the  
13 application and subject to direct spray, so this could result in  
14 some high estimates. This was not what was being studied, so  
15 any bees that were intercepting some of this spray would be  
16 subject to having anomalous concentrations, anomalously high.

17 And then finally, the normalization -- and  
18 this is not just in this particular study, but elsewhere as well  
19 - but the normalization assumes the linear relationship between  
20 the rate of spraying and the rate of accumulation by bees, which  
21 is not necessarily valid. I will talk about this later in case  
22 there are some questions about this. Anyway, those are my  
23 comments. I know that Tom - you also have something?

24 **DR. DANIEL SCHLENK:** Thanks. Dr. Potter?

25 **DR. THOMAS POTTER:** Okay. I certainly concur  
26 with Dr. Schwab. I believe that this study was reasonable and



1 certainly an elegant piece of work, and gets at some very  
2 important questions using a very novel approach.

3 My primary concern about the study is in the  
4 manner of which the dye was applied to the leaves. It certainly  
5 does not necessarily - and its certainly unlikely to mimic how  
6 pesticides are applied or co-applied with a number of different  
7 adjuvants that are blended into the material, including  
8 spreaders and stickers and all sorts of other things.

9 So, that makes this type of study, I guess  
10 somewhat questionable in terms of acting as a firm surrogate for  
11 pesticides and then until such time more work is done, I think  
12 there is considerable uncertainty there in terms of whether this  
13 accurately represents potential exposures or not.

14 **DR. DANIEL SCHLENK:** Does anybody else want to  
15 chime in on this particular question? Okay. Dr. James?

16 **DR. ROSALIND JAMES:** I don't know, Dave, we  
17 talked a little bit about by weight. I mean, and this is  
18 consumption, which the next question is exposure, but again as  
19 we move away from - as we move towards thinking of honey bees as  
20 a surrogate and not just as honey bees, bees come in different  
21 sizes and should the consumption rate be by weight or you have  
22 different -- drones are larger than workers and queens. Do you  
23 have anything you want to add to that?

24 **DR. DAVID TARPY:** This is Dave Tarpy, NC  
25 State. Yeah, that was a comment and a question that I had for  
26 the EPA members yesterday about standardizing it to kind of per



1 milligram of body weight of an individual rather than on a per  
2 bee basis.

3 To kind of get to your point and to your  
4 question, question number 3 about using honey bees as a good  
5 surrogate, that would and I think - maybe I'm wrong here, I  
6 would like other comments on this -- but it seems to me that  
7 that would translate a lot more effectively to other life, to  
8 other castes and to other types of bees to do it on a per  
9 milligram basis. I was going to raise those points in question  
10 number 8 later on. But I think it's applicable here too.

11 **DR. DANIEL SCHLENK:** I see a lot of heads  
12 nodding on that in terms of normalizing per mass it sounds like,  
13 in terms of per bee. Yeah, Dr. Pettis?

14 **DR. JEFF PETTIS:** I agree with what Dr. Tarpy  
15 said. The one exception in honey bee colony would be the queen  
16 in which she, even by her body mass, she's actually processing a  
17 great deal more food than any other individual in the colony.  
18 She is kind of this egg laying machine, thus her nutritional  
19 requirements are extremely high relative to anybody else. So,  
20 body weight wouldn't account for that.

21 **DR. DANIEL SCHLENK:** Dr. Berenbaum?

22 **DR. MAY BERENBAUM:** Am I correct in assuming  
23 that Koch and Weisser's study involved only one race of bee, and  
24 again, there is race variation and size and pursuitness and  
25 other attributes. So, another argument for weight rather than  
26 per bee.



1                   **DR. DANIEL SCHLENK:** So again, let me just  
2 encourage you to get your written comments to Dr. Schwab at  
3 least for this particular question - oh sorry Dr. James, didn't  
4 see you over there.

5                   **DR. ROSALIND JAMES:** Sometimes it is hard to  
6 know where to put the comments, I guess; but also the  
7 consumption rate had been by day. The bee larvae have such a  
8 short life span, that it's often easier to measure it across  
9 their whole life span and with the honey bee, they are only  
10 eating for five days and mostly consumption is done in two days.  
11 There are some early.

12                   Some other possibility would not to be looking  
13 at the consumption per day, but over the lifecycle, I believe  
14 for the larvae. This also would be very applicable to the  
15 solitary bees where they have given them mass provision and then  
16 that is what they eat is that mass provision. Again as I said,  
17 their exposure - whatever day that was collected, that's the  
18 exposure level that they are stuck with unless, of course, you  
19 have degradation over time, but it's a relatively short period  
20 of time that they are feeding on.

21                   So, I did want to suggest not looking at  
22 consumption rate per day for larvae, but at total consumption  
23 for larvae and then the question again about the pollen versus  
24 the nectar, and if there is a difference in pesticide  
25 concentration between pollen and nectar, that ratio could be  
26 very important. Different life stages have different ratios.



1 Your solitary bees are going to be more dependent on pollen than  
2 your worker honey bees as adults. Thank you.

3 **DR. DANIEL SCHLENK:** Sure. Dr. Fefferman?

4 **DR. NINA FEFFERMAN:** So, just to quickly  
5 follow up on that idea, I'm strongly in favor of the consumption  
6 per body weight idea. I also very much like, but I'm not sure  
7 how to integrate with that per body weight idea the idea of  
8 whole mass over your stage feeding, because your weight changes  
9 substantially over the course of being a larva at different  
10 days. So, there are good ways to do it, but one should be  
11 chosen and it should probably be discussed quite carefully how  
12 to do that.

13 **DR. DANIEL SCHLENK:** Yes, Dr. Ostiguy?

14 **DR. NANCY OSTIGUY:** This is Nancy Ostiguy.  
15 This question actually is looking at contact exposure. So, we  
16 may want to look at instead of a milligram per kilogram, a  
17 surface area measurement if we're looking at contact.

18 **DR. DANIEL SCHLENK:** Okay. Jumping the gun a  
19 little bit I guess. The following questions, I'm thinking.  
20 Yes. In terms of diet, yes. Dr. Tarpy?

21 **DR. DAVID TARPY:** To answer Dr. Fefferman's  
22 point, to reconcile those things, one is larval and the other is  
23 adult. So, I think it'd be pretty much impossible, I think  
24 logistically to measure the weight of larvae effectively,  
25 especially as it changes over time. So, it's not a constant.

26 But for adults, they don't grow, so that



1 standardizing by the adult exposure by weight would be possible,  
2 but then the total consumption over the feeding lifetime of the  
3 larvae would be the way I think would do it. Again, I think  
4 this will come up probably again in question number 8.

5 **DR. DANIEL SCHLENK:** You guys have any other  
6 comments from the Panel before I turn it over to the agency?  
7 Okay. Any questions or clarification? Ms. Garber?

8 **MS. KRIS GARBER:** Yes, Kris Garber. Dr.  
9 Schwab, you mentioned that you had some concerns about the  
10 assumption of linear scaling for application rate. I'm curious  
11 if you have an alternate suggestion to the linear assumption?

12 **DR. PAUL SCHWAB:** No, not really. I can't say  
13 that I have something that is superior to that, but that is  
14 definitely an inherent assumption that it is all linear and it  
15 may or may not be.

16 **DR. DANIEL SCHLENK:** Okay. So, we are all  
17 good with (a)? Oh, sorry, Mr. Pistorius has another comment  
18 here.

19 **MR. JENS PISTORIUS:** Sorry, I was a little bit  
20 late. I just wanted to add, we calculated on this approach and  
21 it is actually in line with the HQ data and also the incident  
22 data from exposures, so I think this approach, as it was said  
23 before, it quite conservative.

24 **DR. DANIEL SCHLENK:** Okay. This is for -- Dr.  
25 Schwab, did you get his comments?

26 **DR. PAUL SCHWAB:** Not thoroughly, can you



1 write that up for me?

2 **DR. DANIEL SCHLENK:** All right. Let's go to  
3 (b) if we can, and then we will take a break.

4 **MS. KRIS GARBER:** Kris Garber. Question 4,  
5 part (b) - please comment on the potential utility of the T-REX  
6 upper-bound residue value, i.e. 12 microgram a.i. per bee per 1  
7 pound a.i. per acre for a broader number of arthropod species to  
8 represent contact exposures to honey bees and to other insect  
9 pollinators that are directly sprayed with pesticides.

10 **DR. DANIEL SCHLENK:** Okay. Dr. Schwab?

11 **DR. PAUL SCHWAB:** This approach is sort of a  
12 modeling approach that's being used when there is an absence of  
13 hard data. Obviously, the direct studies of any sort of bees or  
14 pollinators would be preferred, but in the absence of data, this  
15 would be a method that could be used. One of the strengths of  
16 this that the data used in generating T-REX represents actual  
17 measurements on arthropods and are expected to actually reflect  
18 what you might see in honey bees and non-Apis bees.

19 Some of the weaknesses would include that,  
20 although it was based on 14 studies, it was only done on a few  
21 different kinds - actually 2 different categories of pesticides,  
22 the carbamates and the organophosphates. Some of the others  
23 like the neonicotinoids - gee, I probably couldn't pronounce my  
24 own name at the moment - with totally different chemistry  
25 perhaps may not be very well reflected in this model.

26 Let's see, doses and residues are based on



1 normalization. Again, this gets back to the same thing. I  
2 don't have a better way to go, but it does assume that  
3 linearity. That was the sum of my comments at this point.

4 **DR. DANIEL SCHLENK:** Dr. Potter?

5 **DR. THOMAS POTTER:** I will say I agree with  
6 Dr. Schwab in regards to the points and the applicability of the  
7 study. Again to repeat, it would certainly be appropriate to  
8 have measurements on the target organism rather than have to  
9 extrapolate to others. With that said, I'm not sure that the  
10 arthropod data is any more uncertain than the traits for  
11 estimate. They are both, I think, highly uncertain. I think it  
12 is clearly a data gap that could be filled relatively easy  
13 without substantial cost. So, I certainly would want to  
14 emphasize that that be done.

15 I will add perhaps there is another way of  
16 thinking about this in terms of estimating exposure, and that is  
17 simply to take the assumed spray rate, which is 1 pound per acre  
18 or 1 kilogram per hectare, if you do the arithmetic on that,  
19 that becomes 11 micrograms per square centimeter. Hopefully my  
20 arithmetic is working today, but I believe that is correct. So,  
21 that's the application rate for square centimeter. If we look  
22 in the literature, and I did some digging on this and I actually  
23 found this notation. I was pleased, which provided the surface  
24 area of a honey bee.

25 Its these guys, Steve Roberts and John  
26 Harrison, maybe you know them whether they have been working in



1 the field for a number of years, but this was an article  
2 published in the Journal of Experimental Biology in 1999. They  
3 pulled some wings off the bees. That sounded kind of gruesome,  
4 but anyway, they pulled the wings off and measured the surface  
5 area. They came up with 2.4 square centimeters. So, if you  
6 take your 11 micrograms per square centimeter and divide it by  
7 the 2.4, you get essentially a potential contact rate. That is,  
8 in essence, a 45 percent interception rate, if you will, of the  
9 chemical. So, that in some ways, I think it provides, you know,  
10 perhaps another back check in terms of what might be the  
11 reasonableness of this number, this 12 microgram per bee number.

12 Perhaps there are some other folks who are much more familiar  
13 with bees in the room than I am can comment about the surface  
14 area as an issue, but I think that that at least provides a test  
15 of reasonableness to the value.

16 **DR. DANIEL SCHLENK:** Yes, we will go with Dr.  
17 Fefferman first and then Dr. James.

18 **DR. NINA FEFFERMAN:** Sorry, this may be way  
19 too detailed, but in terms of surface area of bees, there are  
20 some regions of bees that they (inaudible) much more frequently  
21 than others, so I don't know if you want to default the entire  
22 surface of your bee unless it's actually sort of an absorption  
23 through cuticle kind of thing. I don't know if bees do that.

24 **DR. DANIEL SCHLENK:** Dr. James?

25 **DR. ROSALIND JAMES:** This is Rosalind James.  
26 Maybe that would be like your upper limit of what they could



1 possibly be exposed to. Unless you are spraying soil, a field  
2 is actually three-dimensional, so that pesticide rate is being  
3 spread over plant leaves and mostly on the upper and not so much  
4 on the lower. And in addition to that, bees are designed to  
5 collect things, and they are full of hairs, and they may  
6 actually collect more liquid than their surface area. So,  
7 contrary to what you would say, I would say the opposite that  
8 they have the surface tension and are made to gather things.  
9 So, that would be a very rough estimate I would say.

10 **DR. THOMAS POTTER:** I will say that, but  
11 again, it's in line with the upper-bound estimate that is being  
12 proposed in the White Paper. So, that's why I put it forward as  
13 to say there is maybe one other way to think about this and that  
14 is indeed looking at surface area and potential for  
15 interception. I agree with all the things you said and  
16 certainly not being a honey bee physiologist, or bee  
17 physiologist at all, I have to default to that. But, you know,  
18 perhaps there is some more thought that can go into this  
19 generalized area, which might help us get some appreciation for  
20 interception rate, essentially is what we are talking about  
21 here.

22 **DR. DANIEL SCHLENK:** Dr. Ostiguy?

23 **DR. NANCY OSTIGUY:** This has come up a little  
24 bit before, but it shows up here very specifically. We are  
25 looking at exposures to honey bees and to other insect  
26 pollinators. As Dr. Berenbaum said yesterday, there are other



1 pollinators than bees. We need to be clear what we are talking  
2 about. So are we talking about honey bees or are we talking  
3 about honey bees and other bee pollinators or are we including  
4 flies, or who are we including?

5 **DR. DANIEL SCHLENK:** Okay. Any other comments  
6 from Panel members? It sounds like there's a proposed check  
7 that's out there with some caveats obviously.

8 **DR. THOMAS POTTER:** I get carried away here.  
9 Tom Potter here. I'm sorry about that. Yes, you know, it's  
10 perhaps another check. It's a factor of two greater than the  
11 proposed value that's been suggested in the White Paper, which  
12 you know - and essentially we're talking about a 100 percent  
13 interception here. So, in that sense, I would think it puts the  
14 value that we are talking about in this case is probably  
15 reasonably conservative.

16 **DR. DANIEL SCHLENK:** Any other comments from  
17 the Panel? Okay. Let me go to the Agency. Any questions or  
18 clarification?

19 **MS. KRIS GARBER:** This is Kris Garber. I just  
20 want to just summarize what I heard back and make sure that I  
21 heard correctly. This is going to pertain to the whole  
22 question. So, it sounds like the proposed value of 2.7  
23 microgram a.i. per bee normalized to 1 pound a.i. per acre is a  
24 reasonable approach and that T-REX wouldn't necessarily provide  
25 an alternative or a better alternative, and that this surface  
26 area approach could be another way to just kind of like another



1 line of evidence to support the use of the 2.7? But you're not  
2 necessarily proposing to use the 2.7 to calculate - I mean,  
3 you're not necessarily proposing to use the surface area  
4 approach to calculate risk quotients.

5 **DR. THOMAS POTTER:** Not at this moment, no.  
6 This is Tom Potter here. Maybe Arthur had initial response  
7 here. I just wanted to leap in on the surface here. I simply  
8 put that as, you know, as another way of thinking about the  
9 problem and helping to define what the maximum exposure. I feel  
10 that whether or not the T-REX 2.7 value or the arthropod 12  
11 value, which one is better - I don't think we have the available  
12 information at this point to say. I would strongly emphasize  
13 that studies need to be done, collecting bees in sprayed fields  
14 and making residue measurements. I believe that that is the  
15 most appropriate path forward here.

16 **DR. DANIEL SCHLENK:** Okay. Are we good? All  
17 right, let's go ahead and break for 15 minutes. Let's try to be  
18 back at 2:30.

19 (WHEREUPON, a recess was taken)

20 Go ahead and get going? We can? All right,  
21 Kris are you going to read 5 in?

22 **MS. KRIS GARBER:** Yes. This is Kris Garber  
23 again. Charge question 5, part A - although bee larvae  
24 typically consume processed foods in the form of royal jelly and  
25 brood food throughout much of their development, they also  
26 consume honey and pollen during the last two days of the



1 uncapped period. Please comment on the proposed use of nectar  
2 and pollen consumption rate of larvae during the last day of the  
3 larval developmental stage. Please include a discussion of the  
4 conservatism, strengths and limitations of this approach as well  
5 as a discussion of how this value may or may not correspond to  
6 data generated from larval toxicity endpoints.

7 **DR. DANIEL SCHLENK:** Okay. Our lead  
8 discussant on that is Dr. Pettis.

9 **DR. JEFF PETTIS:** Yes, Jeff Pettis. I'll lead  
10 with a question that we have for EPA about the data presented  
11 where royal jelly concentrations were graded 100 times lower  
12 than that in food consumed. I just like - in a few minutes when  
13 EPA has a chance to respond - to talk about those studies  
14 whether they've been published, whether they're a single study  
15 or multiple data points. I would like some clarification  
16 because they affect our response.

17 But in general, we agree that unprocessed  
18 nectar and pollen would represent a worst case scenario. If we  
19 accept that, then they should be a conservative estimate by  
20 using them over brood food or royal jelly.

21 As far as relevance to the larval toxicity  
22 endpoints, if you are doing larval assays, most of those are  
23 done with dilute royal jelly and so, the use of nectar and  
24 pollen again would be a conservative estimate and may not lead  
25 directly to larval endpoints. Most of those larval endpoints  
26 are done by spiking the royal jelly or the brood food. So they



1 provide a valid assay tool for looking at larval toxicity  
2 issues.

3 We see no issue with the use of dilute royal  
4 jelly in these larval assays. Some of the larval assay data  
5 that I've seen generated shows that when using brood food or  
6 dilute royal jelly, the adults produced in these assays lived as  
7 long and foraged normally as would workers reared in whole  
8 hives. So, those larval assays are valid, and again back to the  
9 use of nectar and pollen, I think they've produced a  
10 conservative estimate. So, that's just for 5A.

11 **DR. DANIEL SCHLENK:** Dr. Ostiguy?

12 **DR. NANCY OSTIGUY:** I concur.

13 **DR. DANIEL SCHLENK:** Okay. Other panel  
14 members? Yes, Dr. Tarpy?

15 **DR. DAVID TARPY:** This is Dave Tarpy. Jeff, I  
16 know that your lab has experience with these larval bioassays,  
17 but there is a lot of other labs that have slightly different  
18 approaches and in some cases, quite different approaches. So,  
19 how those translate to each other is, I think, something of  
20 consideration. So, is there an inherent assumption here of  
21 always having a control group in these bioassays to make sure  
22 that these things are relativized to different concentrations of  
23 the compound.

24 **DR. JEFF PETTIS:** Jeff Pettis. I guess - I  
25 think there is a need for standardization within these larval  
26 assays. I know there has been some ring testing in the EU to



1 try to standardize and make these larval assays valid. I don't  
2 think we are quite there yet with most of the labs in the U.S.

3 **DR. DANIEL SCHLENK:** Dr. James?

4 **DR. ROSALIND JAMES:** But at the same time,  
5 larval assays are exceedingly important. I mean, so we have to  
6 find a way to do them.

7 **DR. DANIEL SCHLENK:** Mr. Pistorius?

8 **MR. JENS PISTORIUS:** I would like to add to  
9 the comment of Jeff, that it is the same in the EU. I think  
10 most labs are not currently there yet. I have a question, or  
11 maybe a concern or I'm not sure what it is - maybe we will also  
12 discuss it later in the Aupinel section of question 9, but now  
13 when I read this here, I read that the concentration in royal  
14 jelly is in fact 100 lower. I think that somewhere it should be  
15 discussed if we do such a larva toxicity test, but maybe this is  
16 too early to ask. Are we actually using the lower  
17 concentrations, 100 times lower? If you do a risk assessment  
18 refinement, you have residue data in nectar and pollen, do you  
19 then say we go for concentration 100 times lower? Then again, I  
20 apologize if it's not appropriate to ask this at this time.

21 **DR. DANIEL SCHLENK:** Well, I think that's up  
22 to Dr. Pettis if he thinks it appropriate to answer that  
23 particular question with that sort of component. Would you  
24 rather do that now or wait, I guess?

25 **DR. JEFF PETTIS:** I think I would like to  
26 address it - Jeff Pettis. I'm hoping that the idea would be not



1 to use 100 times lower value, but to use more of a worst case  
2 scenario in a larval assay.

3 **MR. JENS PISTORIUS:** Agreed.

4 **DR. DANIEL SCHLENK:** okay. Sounds good. Any  
5 other comments from the Panel for A? Okay. We go to the  
6 Agency. Are you clear with that response?

7 **MS. KRIS GARBER:** Yes, I actually do have one  
8 clarifying question though. I can also address Dr. Pettis's  
9 question about the 100 times factor. So, Dr. James, you have  
10 mentioned earlier that we should consider accounting for dose  
11 over the entire larval life stage. So, would that entail  
12 expanding the consumption rate out to the fourth and fifth days  
13 or all five days?

14 **DR. ROSALIND JAMES:** If I remember right, your  
15 data was actually on the entire food consumed, and then you had  
16 to extrapolate out to a daily rate - right?

17 **MS. KRIS GARBER:** This is Kris Garber. So to  
18 clarify, it was broken out by the first three days and then food  
19 consumption over the fourth and fifth days.

20 **DR. ROSALIND JAMES:** I would say the whole  
21 life stage. The royal jelly for the first three days and then  
22 the pollen and nectar consumed during the fourth and fifth or  
23 out to the seventh, depending on whether you are talking about  
24 workers or drones. I might as well say this here now - you sort  
25 of ruled out royal jelly because it had lower pesticide  
26 concentration in it, but if I remember right, that was based on



1 one study. Is that right?

2 **MS. KRIS GARBER:** So, this gets to Dr.  
3 Pettis's question also. It was based on two studies. The first  
4 one is actually a published study involving dimethoate and  
5 carbofuran. I believe the authors were Davis and Shuel, and  
6 it's cited on appendix 1, so you can confirm that. Actually,  
7 they observed concentrations that were more than 100 times  
8 different. I think it was a factor of 1000? And then the  
9 second study - those results are unpublished. Part of the  
10 results - the analysis was conducted by EPA, and that involved  
11 Imidacloprid. That was actually the basis for the 100X factor.  
12 So, nurse bees were fed concentrations of food that were 100  
13 times more than what showed up in the royal jelly that they were  
14 producing.

15 **DR. DANIEL SCHLENK:** Just hold on a second.  
16 So, Dr. Pettis does that answer your question?

17 **DR. JEFF PETTIS:** That does answer the  
18 question.

19 **DR. DANIEL SCHLENK:** Okay. Great. And then  
20 Dr. Berenbaum, you have another comment?

21 **DR. MAY BERENBAUM:** Just to say that this does  
22 come up in question 10, that the royal jelly estimates are based  
23 on one, but also there are some issues about the relative  
24 nutritional value and contribution, even if discounting the  
25 consumption of royal jelly ignores the relative impact on  
26 growth, but that may be more relative for question 10 right now.



1  
2 **DR. DANIEL SCHLENK:** Let's wait for question  
3 10, yeah, if we can. Any other 5A questions? Any other 5A  
4 comments? Okay. Let's move to the second letter of the  
5 alphabet. Somebody asleep out there.

6 **MS. KRIS GARBER:** This is Kris Garber. Charge  
7 question 5, part B - please comment on the strengths and  
8 limitation of basing the Tier I screen for adult honey bees on  
9 food consumption rates of nectar foraging bees, including a  
10 discussion of the conservatism of this approach and how it  
11 relates to other types of worker bees and castes.

12 **DR. DANIEL SCHLENK:** Okay. Dr. Pettis?

13 **DR. JEFF PETTIS:** In general, I think the use  
14 of nectar consumption is generally appropriate, but may not be  
15 reflective of the age of the bees, which are often tested in  
16 Tier I. Tier I testing could use either randomly aged bees or  
17 young bees, and especially if they are young bees or newly  
18 emerged bees, then their diet normally would contain high  
19 amounts of pollen or pollen intake. So the use of just nectar  
20 consumption information may not be as appropriate as we would  
21 like.

22 As far as relative to worker bees and other  
23 castes, as was brought up earlier by Dr. Tarpy, using weight  
24 might be a better estimate accounting for caste differences with  
25 the exception of the queen as I have already pointed out. But I  
26 do think that in general, worker bees - obviously they are



1 easier to manipulate, they're available year round, so their use  
2 in Tier 1 screening is appropriate. With the exception of the  
3 pollen intake for young bees, it may not be reflective of the  
4 nature progression of the worker bee development. So, if you  
5 don't include some protein intake into the Tier I testing, which  
6 I don't think is normally the case, then I think we are missing  
7 an important component of their lifespan. That's all I have for  
8 that.

9 **DR. DANIEL SCHLENK:** Okay. Dr. Ostiguy?

10 **DR. NANCY OSTIGUY:** I concur.

11 **DR. DANIEL SCHLENK:** Okay. Other Panel  
12 members? Dr. James?

13 **DR. ROSALIND JAMES:** Just to reiterate about  
14 using honey bees as a surrogate for other bees, the non-Apis  
15 bees, all of the bees are going to probably eat pollen in  
16 addition to nectar. You can think of perhaps, every bee as a  
17 queen in the solitary bee. So, just one more argument for  
18 including pollen in the diet. It really maybe only matters if  
19 you have a difference in pesticide exposure between pollen and  
20 nectar.

21 **DR. DANIEL SCHLENK:** Anyone else. Yes, Dr.  
22 Hunt?

23 **DR. GREG HUNT:** I would just like to say I  
24 agree that including pollen is important because if you don't,  
25 it's an artificially high stress on the bee.

26 **DR. DANIEL SCHLENK:** Okay. Dr. Pettis?



1                   **DR. JEFF PETTIS:** And just one other thought  
2 that occurred to me during the discussion was that with some of  
3 the systemics, the residue levels in pollen are generally higher  
4 than they are nectar. I think that has already been taken into  
5 account, but again, pollen consumption is normal for young bees.

6  
7                   **DR. DANIEL SCHLENK:** Anyone else? Okay. Ms.  
8 Garber is that - any questions or clarification?

9                   **MS. KRIS GARBER:** I do have one point of  
10 clarification. I know that it's often difficult to separate out  
11 the exposure and effects components of the process, but I just  
12 want to make sure that I understand that the responses are  
13 specific to the exposure side so the consumption - just make  
14 sure that we're not talking about the toxicity tests or the  
15 conduct of the toxicity test. Is that correct?

16                   **DR. DANIEL SCHLENK:** Dr. Pettis?

17                   **DR. JEFF PETTIS:** Could you repeat the -  
18 because I'm not - go ahead.

19                   **MS. KRIS GARBER:** Okay. Thank you. Kris  
20 Garber again. So, the focus of this question is on the  
21 consumptions rates that we would use to estimate exposure in the  
22 Tier I analysis, and I know there is some overlap with how it  
23 may be related to the toxicity side of the risk quotient  
24 equation. I just want to make sure that the comments aren't  
25 focused on giving us advice on how to conduct the toxicity tests  
26 themselves.



1                   **DR. JEFF PETTIS:** Your point is well taken. I  
2 think there was some overlap in that, so yeah, you're right. If  
3 you're trying to look at just exposure, then we have different -  
4 some of the comments about pollen consumption and all were maybe  
5 not as appropriate as they might have been.

6                   **DR. DANIEL SCHLENK:** Okay, Dr. James?

7                   **DR. ROSALIND JAMES:** Along those lines, it  
8 mainly matters if there is a difference in past site  
9 concentration in the pollen versus the nectar. And there's a  
10 question that still seems to be hanging in the air. So, until  
11 there is more hard data on that, you do sort of need to keep the  
12 pollen and nectar ratio in mind when you are doing the  
13 calculations I think.

14                  **DR. DANIEL SCHLENK:** Dr. Ostiguy?

15                  **DR. NANCY OSTIGUY:** That actually brings up  
16 something that I have been thinking about and that is an area  
17 where we have a data gap. Half lives of our pesticides are  
18 calculated usually soil, water, et cetera, we don't necessarily  
19 know what the half life would be in the different matrices  
20 within the hive. That to me is a data gap. For us to be able  
21 to actually then extrapolate with our models and say yes, that's  
22 an appropriate exposure.

23                  **DR. DANIEL SCHLENK:** Dr. Berenbaum?

24                  **DR. MAY BERENBAUM:** And in terms of exposure  
25 too, one of the issues of using limited amount of data to  
26 estimate pesticide concentrations particularly when they are



1 older studies is that today's chemistries are very different  
2 from old chemistries, so we really don't know whether they're  
3 comparable. So, that is why it's an issue of whether it would  
4 be another data gap in terms of estimating the degree of  
5 exposure through foods like royal jelly. We just don't have  
6 those data.

7 **DR. DANIEL SCHLENK:** Okay. Any other comments  
8 for 5B? Are you good with 5B? Yeah? Okay. The Agency is  
9 nodding their heads. All right, let's read 5C into the record  
10 then.

11 **MS. KRIS GARBER:** Kris Garber. Charge  
12 question 5C - please comment on the assumption that exposures  
13 through consumption of nectar and pollen are conservatively  
14 representations of potential exposures through consumption of  
15 honey, bee bread, brood food and royal jelly, all of which  
16 represent processed foods.

17 **DR. DANIEL SCHLENK:** Dr. Pettis?

18 **DR. JEFF PETTIS:** Our general assumption is  
19 that we agree with this that in fact, nectar and pollen would  
20 represent a worst case scenario or more so than processed food.  
21 As example, you bring out the royal jelly - it's a processed  
22 food - goes through the honey bee and there are processed  
23 intraglandular secretions, so even though bee bread and other  
24 stored products would undergo some degradation, so yes, we agree  
25 that it's the worst case scenario.

26 **DR. DANIEL SCHLENK:** Okay. Dr. Ostiguy?



1                   **DR. NANCY OSTIGUY:** This is actually one  
2 situation where our extrapolation to native bees might not be as  
3 far because they're actually provisioned as larva with pollen  
4 and potentially a little bit of nectar. So, they're not getting  
5 brood food or royal jelly.

6                   **DR. DANIEL SCHLENK:** Okay. Dr. Berenbaum?

7                   **DR. MAY BERENBAUM:** Does anyone actually know  
8 the degree to which processing effects pesticide concentrations?  
9 I know of only one case in the literature, and it's degradation  
10 of phytochemicals in nectar or honey as it cures, but I don't  
11 know of any studies that have shown degradation of pesticides.

12                   **DR. DANIEL SCHLENK:** Sounds like a data gap.  
13 Okay. I guess we want to put that in the response as a data  
14 gap. Yeah, Dr. James?

15                   **DR. ROSALIND JAMES:** So, this is Rosalind  
16 James. May, would you say it's a reasonable assumption that it  
17 the concentration would not increase with processing?

18                   **DR. MAY BERENBAUM:** Well, it increases in  
19 nectar. It increases when you go from nectar to honey because  
20 it's concentration. It goes from, you know 80 percent water to  
21 18 percent water, so that is an increase in terms of what  
22 happens as it is stored in the hive. I don't think anybody  
23 knows. It is not going to increase, presumably, although there  
24 are some pesticides like organophosphates that can be  
25 bio-activated. Whether or not they are in honey or bee bread, I  
26 don't know if anybody would know. I'm not saying it's a good or



1 bad - it's not an adequate proxy, I just don't know.

2 **DR. DANIEL SCHLENK:** Dr. Sandy?

3 **DR. MARTHA SANDY:** I think another data gap is  
4 we don't know what the degradation or metabolic products,  
5 whether they might be more toxic than the parent compound to the  
6 bees.

7 **DR. DANIEL SCHLENK:** Yes, Mr. Pistorius?

8 **MR. JENS PISTORIUS:** I don't think that  
9 although in the first moments on logic, when you go from nectar  
10 to honey that you would have a concentration of the residues,  
11 but I think data that we have and that were generated in EU has  
12 shown that it's actually vice versa, that you have a lower  
13 concentration in honey than nectar. And does data, for  
14 instance, also opinion which undermines it?

15 **DR. DANIEL SCHLENK:** Dr. Berenbaum?

16 **DR. MAY BERENBAUM:** Is that published?

17 **MR. JENS PISTORIUS:** Yes, those are published  
18 data, for instance, in the Scientific Opinion 2012. There is a  
19 list of data that was used.

20 **DR. DANIEL SCHLENK:** Mr. Pistorius. Dr.  
21 Fefferman?

22 **DR. NINA FEFFERMAN:** Sorry, just for my own  
23 clarification, is that given the assumed half life of the active  
24 agents in the pesticide over that duration of time that it takes  
25 to reduce to the honey irrespective of honey reduction, or is it  
26 the action of reducing the nectar to the honey that you think is



1 doing that?

2 **MR. JENS PISTORIUS:** I'm not quite sure what  
3 is the reason for it? There has been data from (inaudible) who  
4 have shown that actually when the bees incorporate nectar and  
5 then they have it in the honey they still make themselves that  
6 actually after a few hours, the concentration is lower. So, the  
7 bee seems to absorb a little bit, but that's only one process.  
8 There seems to be another process that over time, and here we  
9 only have data - we have measurements of nectar directly when  
10 the foragers come back home, and then nectar in the hive and  
11 then this nectar to honey again. You can see that the  
12 concentrations get lower and lower, but I'm not quite sure what  
13 the exact process is behind it.

14 **DR. DANIEL SCHLENK:** So, that would support  
15 conservation in terms of consumption of nectar and pollen,  
16 right? Yes.

17 **DR. MAY BERENBAUM:** May Berenbaum. Can we  
18 include that in our - can we include that reference in our.

19 **DR. DANIEL SCHLENK:** Definitely. I would  
20 think that would support exactly what Dr. Pettis initially said,  
21 yeah. Any other comments? Okay. Dr. Pettis?

22 **DR. JEFF PETTIS:** I just would like to ask the  
23 group if they kind of agree with the things that are processed,  
24 the royal jelly or brood food, if there is general consensus  
25 that those values would be lower. It sounds like with honey, it  
26 could also be lower, but with pollen, there is a lot of level of



1       uncertainty as far as stored pollen versus bee bread.

2                   **DR. DANIEL SCHLENK:**   Okay.   Everyone shake  
3       their heads.   It looks good.   Oh, question mark?   No?   Okay.  
4       Yeah, a lot of uncertainty there, it sounds like.   Any other  
5       comments for 5C?   Okay.   Let me go Kristina - do you have any  
6       questions or clarification?

7                   **MS. KRIS GARBER:**   No, we're good.

8                   **DR. DANIEL SCHLENK:**   Okay.   Great.   All right,  
9       let's move on to charge question 6.   Going to read letter A into  
10      the record?

11                  **MS. KRIS GARBER:**   Kris Garber.   Charge  
12      question 6, part A, foliar spray - please comment on the  
13      analysis presented in Section 3.1.1.1, with a focus on the  
14      extent to which the T-REX tall grass upper-bound residue may  
15      serve as an adequate surrogate to represent upper-bound  
16      pesticide concentrations in pollen and nectar of flowers that  
17      are directly sprayed with pesticides.

18                  **DR. DANIEL SCHLENK:**   Okay.   Our lead  
19      discussant on that is Dr. Schwab.

20                  **DR. PAUL SCHWAB:**   Okay.   Paul Schwab.   The use  
21      of the T-REX model is necessitated due to the lack of data  
22      directly available on this subject.   So, clearly data would be  
23      better, but when we look at the strengths of this, both in terms  
24      of the nectar and the pollen from the point of view of the  
25      nectar, T-REX is used currently by EPA.   This is something that  
26      has been done for a while.   It was based on actual experimental



1 data as we spoke on the last question. The model actually uses  
2 very few input parameters, which helps in terms of its  
3 simplicity of use. It was developed with a large quantity of  
4 field data. The upper limit values are the upper-bound on  
5 residues from the actual measurements.

6 The empirical data for nectar concentrations  
7 were compiled to evaluate the upper boundaries of T-REX and then  
8 specifically there were seven studies that were used. And, tall  
9 grass was chosen because it is three times higher than the  
10 upper-bound value in the empirical data. So, they were choosing  
11 from different species that were available and felt that the  
12 tall grass would be the best to sort of match the conservative  
13 approach that was being sought, and yet have as few as possible,  
14 if any, values of the empirical data exceed the tall grass  
15 value.

16 From a pollen point of view, 11 studies for  
17 direct spray were used in developing for T-REX. The values that  
18 were compared to the empirical value show that the T-REX for  
19 tall grass is two times higher than the highest measure, so it  
20 is conservative. Twenty three actual data point were used in  
21 the development of this set, and the 110 milligrams per kilogram  
22 per pound of active ingredients per acre should be protected  
23 then.

24 Some of the weaknesses would include that  
25 clearly no actual nectar or pollen data were used, so this is a  
26 very strong weakness, if that makes sense. Normalize to 1 pound



1 per acre assumes a linear change, again sorry. The model does  
2 not contain actual values for pollen nectar from flowers that  
3 were directly sprayed. The highest empirical values correspond  
4 to a mean measurement of pesticides in nectar not a maximum.  
5 Therefore, the comparisons were made to T-REX as not an  
6 upper-bound really - this is just a matter of the way it was  
7 developed, really doesn't have to do with whether or not it's  
8 conservative. And, crop specific factors really would be  
9 difficult to account for under these circumstances.

10 **DR. DANIEL SCHLENK:** Dr. Pettis?

11 **DR. JEFF PETTIS:** I just have one additional  
12 comment or weakness. The T-REX model may in fact be  
13 conservative, but only if active ingredients are tested. They  
14 may not be predictive of the actual residues of the formulated  
15 product. So, if the formulated product was used, it would  
16 change the movement, penetration and longevity of the active  
17 ingredient in the field. So, that's a weakness of just using  
18 the a.i., the active ingredient.

19 **DR. DANIEL SCHLENK:** Dr. Potter?

20 **DR. THOMAS POTTER:** You know, I generally  
21 concur with both Dr. Schwab and Dr. Pettis. I think tall grass  
22 appears to be conservative, but I am not convinced that we have  
23 the data to determine whether the value is conservative or not.

24  
25 There is an appearance of conservatism, but  
26 the reality is -- and I've made this comment several times



1 during the meeting in the form of questions - we have apparently  
2 a paucity of data, actual residue measurements on nectar and  
3 pollen with which to compare to this estimate.

4 I wanted to make one comment about one of the  
5 papers, peer review papers that was used in the data  
6 compilation. That was the paper by Choudhary and Sharma. I  
7 took a quick read through it, and I think this is one of ones  
8 where maybe a little more peer review would have helped.

9 I had some serious questions after I read  
10 through that paper. I still am puzzled as to how they came up  
11 with the detection limits that they reported. With regard to  
12 study design, the spray was applied when 50 percent of the  
13 flowers were open. There is no accounting for flowers opening  
14 after the spray was applied. The pollen and nectar were  
15 collected from bees that in theory were foraging in their crops,  
16 that they had treated, but there is no way of knowing that.

17 So, we have a data quality problem in terms of  
18 study design. With that said, I think our subset of data that  
19 we have that it is reliable with which to compare to this T-REX  
20 value was even smaller unfortunately. So, I just wanted to make  
21 the comment about that and again, beat on this idea that we need  
22 a data call-in, need to get that information compiled, and  
23 that's going to, I think, give his whole risk assessment process  
24 in terms of dietary exposure a whole lot more credibility.

25 One final point about nectar, because it's a  
26 liquid, was that it's roughly a 30 percent sugar solution.



1 Pesticides do dissolve in it. Therefore, the solubility of  
2 pesticide active ingredients in the nectar ought to be  
3 considered in terms of assessing potential exposures. I think  
4 if we go through that process, we might gain some further  
5 insight into how conservative our estimates are. Again, I  
6 strongly emphasize we need to call in data, we need more  
7 information in order to be able to determine whether T-REX is an  
8 appropriate exposure estimate or not. Secondly, we perhaps  
9 ought to - in the case of nectar, which appears to be the  
10 primary exposure pathway, we need to take into account the  
11 physical and chemical properties of the active ingredients in  
12 terms of ultimately deriving our exposure estimates.

13 **DR. DANIEL SCHLENK:** Any other panel members  
14 want to weigh in on this one? Dr. Hunt?

15 **DR. GREG HUNT:** Yeah, I don't know if this  
16 data is relevant to Tom's question, but the managed pollinator,  
17 coordinate agricultural project has sentinel apiaries and in six  
18 different states, I believe, throughout the country and  
19 collecting all kinds of pesticide residue data on bee-collected  
20 pollen and wax and bees, and there are some other published  
21 studies that are looking at extant samples, so that may help to  
22 identify maximum levels that are seen.

23 **DR. DANIEL SCHLENK:** Would you provide that  
24 information to Dr. Schwab, I guess, for his comments so that he  
25 can get that in there? Yes, Mr. Pistorius?

26 **MR. JENS PISTORIUS:** Again, from the EFSA



1 opinion or the EFSA opinion site that there are a few more  
2 studies that undermine some of the residue data that are  
3 mentioned here in Table 3 of the White Paper, so I completely  
4 agree and offer to send those data to compile them.

5 **DR. DANIEL SCHLENK:** Thanks. Anyone else to  
6 comment? Okay. Go to Ms. Garber, any questions or  
7 clarification? No? Okay. Read in the letter that follows A.

8 **MS. KRIS GARBER:** Kris Garber. Charge  
9 question 6, part B, soil application - please comment on the  
10 analysis presented in Section 3.1.2, with a focus on the extent  
11 to which the Briggs' model may generate estimates of pesticide  
12 exposure in plant stems that can represent upper-bound pesticide  
13 concentrations in pollen and nectar of flowers.

14 **DR. DANIEL SCHLENK:** Dr. Schwab.

15 **DR. PAUL SCHWAB:** Okay. Looking at this from  
16 the point of view of the strengths. This has been used in a  
17 couple of different approaches. It's used in the PRZM model.  
18 It has also been selected by United Kingdom Environmental Agency  
19 as coming closest to estimating empirical data. All the  
20 pesticides that were used in this model were nonionic. That's  
21 not necessarily a strength, but it does relate to an awful lot  
22 of the pesticides in question.

23 The range in the Log Kow values that were used  
24 in this pretty well encompasses the range of Kows of various  
25 pesticides that we discussed thus far. The original model was  
26 based only on the Log Kow and regression analysis of



1 experimental data, which made it fairly simple. Ryan et al.  
2 modified this and added three soil parameters to make the model  
3 more soil specific. They added the soil bulk density, so a  
4 water content, fraction of organic content in the soil. This it  
5 makes it then possible because the way that the model was  
6 developed was out of solution. This makes it possible then to  
7 do it out of the soil. It does require having these parameters,  
8 however.

9 It was tested against experimental data that  
10 is by Briggs' on five chemicals. Modeling was conservative to  
11 maximize the concentrations in solution. Now the predicted data  
12 are within an order of magnitude to be experimental, but in the  
13 White Paper, a development was used to get the 95th percentile  
14 upper-bound estimates for the transportation stream  
15 concentration factors. This was discussed yesterday.

16 Some of the weaknesses are that it is indeed a  
17 model. It does only examine nonionic compounds, so actually the  
18 mechanism of uptake could be completely different for ionizing  
19 species. The insecticides that were examined were carbamates  
20 and phenylureas, and based on only one kind of plant. This is  
21 definitely a drawback.

22 The soil parameters that were used in the Ryan  
23 modification may or may not be known. Most particularly, the  
24 soil moisture content will be a huge variable. Bulk density  
25 needs to be known, but along with the fraction of the organic  
26 carbamate is very much spacially dependent both in terms of the



1 horizontal and vertical directions. So, this can actually add  
2 some uncertainty to the whole process. I mean, it's a necessary  
3 step, but it doesn't really tighten up the model very much.

4 Now the model is used for stem concentrations  
5 in the transportation stream and not for pollen or for nectar.  
6 So, it sort of gets back to the same problem we had with T-REX,  
7 in that we are examining one thing and extrapolating to another  
8 in terms of plant tissues.

9 In testing the model against five chemicals  
10 with experimental data, various assumptions had to be made  
11 because this was not really using - well apparently these were  
12 unknown. So, there are multiple other assumptions. So, this  
13 would get into some uncertainty again.

14 Not all the values predicted by Briggs' model  
15 were greater than the experimental values. This is why they  
16 went to the 95th percentile method, but even when this was done  
17 to try to gather the upper bound, still not all the data were  
18 above the empirical data, so it's not really truly conservative.

19  
20 I would like to add, some discussion has been  
21 forward both within the White Paper itself as well as other  
22 comments that this particular relationship is somewhat dated,  
23 somewhat limited. But, I would like to throw in that it was  
24 modified, reexamined and other data were put together by  
25 Burghardt and Schonherr in 1998 added another 10 or 15 years or  
26 so to updating this. These were a much broader array of



1 chemicals used. Also a different plant species was used. So,  
2 this could really add to the dataset and help strengthen the  
3 model. That reference will be included in my comments.

4 And finally, we need to put an upward bound on  
5 the water concentration of the pesticide determined by  
6 solubility of the compound in question. It kind of gets back to  
7 sort of the nectar question that Tom was talking about just a  
8 moment ago, in that we can't assume that we can get to just any  
9 water soluble or any water concentration, but that we must  
10 identify an upper bound and make sure that it's not exceeded  
11 because of the physical properties of these compounds.

12 **DR. DANIEL SCHLENK:** Thank you. Dr. Pettis?

13 **DR. JEFF PETTIS:** I agree with the comments of  
14 Dr. Schwab. I just want to elaborate a bit on one more aspect,  
15 and that is the use of stem data to extrapolate to nectar and  
16 pollen. I would agree in principle that it's a conservative  
17 estimate, and there are a few published studies out there, which  
18 examine stem, pollen and nectar, and almost all of those show  
19 that there are higher concentrations in the stem or leaf. So, I  
20 would agree that in general, that's a valid assumption. One  
21 caveat is that plant by plant could be quite different. So,  
22 trying to extrapolate to all plants from hardwood or few crop  
23 plants may not be valid. So it could be a crop-by-crop  
24 difference there.

25 **DR. DANIEL SCHLENK:** Okay. Thanks. Dr.  
26 Potter?



1                   **DR. THOMAS POTTER:** I concur. I think that  
2 both Dr. Schwab and Dr. Pettis have, I think, hit on the points.

3 I generally agree that Briggs is a good first cut. I want to  
4 emphasize that there are substantial uncertainties when that  
5 model is applied to charge species and that needs to be taken  
6 into account. Indeed, there are many active ingredients,  
7 particularly in the herbicide realm these days, that are  
8 charged, in fact I think increasingly, we will see more of that.

9 So, that's a major data gap that needs to be filled, not only  
10 for this risk assessment, but for the use of this same equation  
11 and many other pesticide fate and transport models. It has been  
12 propagated through the system, shall we say. So, there is  
13 substantial effort needed to update that model, particular for  
14 charge species.

15                   **DR. DANIEL SCHLENK:** Any other comments?  
16 Okay. Ms. Garber, any questions or clarification?

17                   **MS. KRIS GARBER:** No, we're good thank you.

18                   **DR. DANIEL SCHLENK:** Great.

19                   **DR. PAUL SCHWAB:** Dr. Schlenk, before we move  
20 on, may I request that we reverse the order of discussion on the  
21 next two?

22                   **DR. DANIEL SCHLENK:** Sure.

23                   **DR. PAUL SCHWAB:** I think that it would be a  
24 more natural discussion to talk about the 1 milligram per  
25 kilogram value before we compare that the Briggs' model. Would  
26 that be okay?



1                   **DR. DANIEL SCHLENK:** So you'd like to go with  
2 D first before C?

3                   **DR. PAUL SCHWAB:** Yes.

4                   **DR. DANIEL SCHLENK:** Okay. Sure. Go ahead  
5 and read D in, great.

6                   **MS. KRIS GARBER:** Kris Garber. Charge  
7 question 6, part D, seed treatments - please comment on the  
8 analysis presented in Section 3.1.3, including a discussion of  
9 the strengths and limitations of the use of 1 milligram a.i. per  
10 kilogram value as an upper-bound concentration for pollen and  
11 nectar of seed-treated crops.

12                  **DR. DANIEL SCHLENK:** Thanks. Dr. Schwab?

13                  **DR. PAUL SCHWAB:** Okay. This particular  
14 approach is addressing section 3.2.2 of the white paper, and  
15 quoting from that, it says that if pesticide specific residues  
16 in pollen and nectar are not available, a screening value of 1  
17 milligram a.i. per kilogram plant matrix is the assumed  
18 exposure. This is from EPPO 2010. This is based on the upper  
19 limit value from empirical data of pesticide concentrations and  
20 different plant parts. The strengths of this would be that it  
21 is stated that the 1 mg per kg value tends to be higher than the  
22 reported concentrations. Most pesticides tend to be about two  
23 orders of magnitude below this particular value.

24                   Obviously, a weakness of this is that because  
25 there are no data that this is just simply - it's not exactly  
26 random, but it is selecting a value rather than depending on



1 data. We sort of visited this concept before and this is a real  
2 obvious problem here.

3 The value may not be completely protective.  
4 The value really takes into account no real aspects of the real  
5 situations that are going on in the field, but that's why  
6 there's no data.

7 **DR. DANIEL SCHLENK:** Okay. Dr. Pettis?

8 **DR. JEFF PETTIS:** I concur.

9 **DR. DANIEL SCHLENK:** And Dr. Potter?

10 **DR. THOMAS POTTER:** I concur.

11 **DR. DANIEL SCHLENK:** Wow. Any other comments?

12 Okay. Ms. Garber? Are you good with that one?

13 **MS. KRIS GARBER:** Good. Thank you.

14 **DR. DANIEL SCHLENK:** So, then, you want to  
15 read in C next, is that correct? Okay.

16 **MS. KRIS GARBER:** Charge question 6C, soil  
17 application - please discuss the relative strengths and  
18 limitations of the 1 milligram a.i. per kilogram value and the  
19 soil uptake model (the Briggs' model) proposed in the white  
20 paper as Tier I screens, including consideration of the extent  
21 to which this method may generate conservative Tier I estimates  
22 of dietary exposures to bees. Does the Panel conclude that the  
23 one approach may be better suited to specific types of  
24 assessment scenarios? If so, please elaborate. Alternatively,  
25 if both approaches are equally suited for a Tier I screen,  
26 please provide guidance on how to best capture variability and



1       uncertainty in the exposure estimates using the two approaches.

2                   **DR. DANIEL SCHLENK:**   And Dr. Schwab?

3                   **DR. PAUL SCHWAB:**   Okay.   There were three  
4       parts to this.   One is to compare the two and the other is to  
5       try to decide if one is better suited to specific scenarios than  
6       the other, and finally then to provide guidance on capturing  
7       variability and uncertainty.

8                   We just reviewed the Briggs' model as well as  
9       the 1 milligram per kilogram level.   That won't be repeated  
10      here, but EPPO approach is that if all positions - assuming that  
11      nothing is known about the soil, the plants or the pesticides -  
12      and this approach assumes that any attempt to estimate pesticide  
13      residues offers no more reliability or even unreliability than  
14      simply assigning a value.   The Briggs' model makes an attempt to  
15      try to integrate information about the soils and the chemistry  
16      of the chemical in the plants, to try to make a projection of  
17      the concentrations.

18                  So, in this way, the Briggs' model might be a  
19      more suitable way to go.   Unfortunately, it also has its  
20      uncertainties because of limitations of the number of chemicals  
21      that were involved and development of the model as well as the  
22      limited number of plant species.   So this is clearly a problem.

23  
24                  In terms of one approach being better than the  
25      other for a given scenario, I would assume then that the Briggs'  
26      model would be best used for those plant species that are



1 closest to the - as modified also by Burghardt and Schonherr -  
2 best suited to those plant species that were used in the  
3 development of the model. We would have to investigate more,  
4 more information to see where this model broke down. So, just  
5 assigning a single value would be best.

6 Then finally in terms of providing guidance on  
7 how to capture variability and uncertainty, there is so much  
8 uncertainty and it's really hard to pin this down.

9 **DR. DANIEL SCHLENK:** Dr. Pettis?

10 **DR. JEFF PETTIS:** I concur with the comments  
11 of Dr. Schwab.

12 **DR. DANIEL SCHLENK:** Dr. Potter?

13 **DR. THOMAS POTTER:** I concur.

14 **DR. DANIEL SCHLENK:** Okay. Anyone else on the  
15 Panel? Ms. Garber, you clear with that one?

16 **MS. KRIS GARBER:** Clear, thank you.

17 **DR. DANIEL SCHLENK:** Great. Okay. So, last -  
18 let's get back to letter E, read that into the record please.

19 **MS. KRIS GARBER:** Kris Garber, charge question  
20 6E - please comment on other approaches or data that should be  
21 considered for estimating upper-bound estimates of pesticide  
22 residues in pollen and nectar as a Tier I screening-level  
23 assessment for pesticides applied via foliar spray, soil  
24 application or seed treatment.

25 **DR. DANIEL SCHLENK:** And Dr. Schwab?

26 **DR. PAUL SCHWAB:** Well, just sort of quickly



1 reviewing the methods that have been described in terms of  
2 trying to get a handle on these values, we have direct  
3 measurements of pesticides and questions on the very specific  
4 plant tissues of interest, whether they are nectar or pollen or  
5 whatever. Secondly, there would be direct measurement of the  
6 pesticide in question and other plant issues such as stems and  
7 leaves and so on.

8 Third, direct measurement of surrogate  
9 pesticides in various plant tissues, then we have modeling  
10 approaches after foliar spray, such as the T-REX modeling,  
11 pesticide uptake from the soil, the Briggs'/Ryan/Burghardt  
12 approach to this. And then finally the sixth one that we looked  
13 at, assigning a default value with no other method is available.

14  
15 This is sort of an exhaustive list. There  
16 might be some other methods. There might be some other modeling  
17 methods, some purely mathematical kinds of approaches that might  
18 be taken, but the way I - at least in my own limited perspective  
19 on this would see this as just sort of tweaking what we already  
20 have. Other approaches might not be all that truly different.  
21 Some may disagree.

22 **DR. DANIEL SCHLENK:** Dr. Pettis?

23 **DR. JEFF PETTIS:** I agree. I guess I'd just  
24 like to emphasize that, yeah, we need direct measurements and  
25 real data, and maybe some of this data may be held by the  
26 registrants and could be useful in the evaluation process.



1                   **DR. DANIEL SCHLENK:** And Dr. Potter?

2                   **DR. THOMAS POTTER:** I will throw this out  
3 again that I think that perhaps an upper-bound on nectar has to  
4 do with the solubility of the chemical in the nectar solutions.  
5 I'm not aware of any systematic study looking at solubility of  
6 organic chemicals in nectar, but that's something that can be  
7 relatively easily rectified, and perhaps would be a useful data  
8 gap to fill.

9                   **DR. DANIEL SCHLENK:** Okay. Any other Panel  
10 members? Okay. Ms. Garber, are you good with question 6?

11                   **MS. KRIS GARBER:** Yes we are, thank you.

12                   **DR. DANIEL SCHLENK:** Okay. So, we are  
13 actually doing really well in terms of our schedule. Would I  
14 would like to do is go ahead and finish up the exposure side  
15 today. Maybe we'll take break after that and start the effects  
16 side tomorrow. So, the last exposure question really is 7. I  
17 know you guys haven't met together, but apparently, everybody  
18 has an answer, so we'll go the traditional route. You guys can  
19 go through that. You got it? Okay. So, if we can go ahead and  
20 read 7A into the record. It's way down there. Yeah, I think  
21 it's the next page perhaps - there it is.

22                   **MS. CHRISTINA WENDEL:** Christina Wendel, 7A -  
23 please comment on the strengths and limitations of basing the  
24 Tier I exposure method on contact and diet. Does the Panel  
25 agree that for the majority of pesticide applications, the  
26 primary exposure routes for bees will be represented by contact



1 and diet?

2 **DR. DANIEL SCHLENK:** And Dr. Potter, you are  
3 lead on this one.

4 **DR. THOMAS POTTER:** Okay. And the way this  
5 question seems to broken out, I think we are going to have to  
6 have a short answer to the beginning and then we'll address the  
7 individual exposure pathways down in the subsequent questions.

8 **DR. DANIEL SCHLENK:** Okay.

9 **DR. THOMAS POTTER:** First and foremost, I  
10 would simply say that diet and direct contact are likely  
11 dominant exposure pathways. However, other exposure pathways  
12 have the potential to be quantitatively significant and should  
13 therefore be considered.

14 **DR. DANIEL SCHLENK:** Okay.

15 **DR. THOMAS POTTER:** And we've covered some of  
16 that earlier when we talked about the conceptual model, and we  
17 will get to some of the other issues on that further.

18 **DR. DANIEL SCHLENK:** Okay. Dr. Hunt, I have  
19 you as your first associate.

20 **DR. GREG HUNT:** I don't have anything to add.

21  
22 **DR. DANIEL SCHLENK:** And Dr. Pettis?

23 **DR. JEFF PETTIS:** Just briefly, I agree that  
24 contact and diet may be the primary routes, and that may vary  
25 contact over diet or diet over contact by chemical type. Then,  
26 I don't think we should discount these other routes of exposure



1 yet. We will get to those.

2 **DR. DANIEL SCHLENK:** Other Panel members?

3 Okay. I will go to the Agency. Are we clear with the answers  
4 for 7A? Okay. So let's go to the letter that precedes C.

5 **MS. CHRISTINA WENDEL:** Question 7, subpart  
6 (b), dust - if the panel believes that this exposure route  
7 should be quantitatively included in the Tier I exposure method,  
8 excuse me, Christina Wendel -- for the relevant application  
9 types i.e., seed treatment, please discuss the data that may be  
10 needed to address the exposure route quantitatively.

11 **DR. DANIEL SCHLENK:** And Dr. Potter?

12 **DR. THOMAS POTTER:** Okay. Well first, I would  
13 like to propose a slight modification to the question here,  
14 which is we don't restrict it to seed treatments. Is that  
15 acceptable?

16 **DR. DANIEL SCHLENK:** I think as long as you  
17 state that's how you answer the question, then you can put that  
18 in there.

19 **DR. THOMAS POTTER:** Okay. And I say that  
20 because I think that there are basically two sources of  
21 pesticide laden dust that may be present in the environment of  
22 the bees. Certainly, emissions from seed treatments have been  
23 widely discussed. I believe Jens Pistorius is going to say  
24 something about that, so I won't get into detail on that. I  
25 think the data strongly indicates that it should be considered.  
26



1           Ideally, we should have information that  
2     quantifies those admissions. You know, we've heard they exist,  
3     but not being a person who has conducted research in this area,  
4     I'm hard pressed to know what the emission rates are from  
5     planters.

6           We've also heard that there are a number of  
7     mitigation measures that can be applied in modified planters and  
8     reducing emissions. Certainly, it's important and relevant that  
9     we get quantitative information about how effective those  
10    practices may or may not be. So, with regard to the seed  
11    coating, I think we again, the theme is we need more data and we  
12    are greedy for data here.

13          The second part of the dust issue, which I'd  
14    like to raise, which hasn't come up in the discussion and didn't  
15    surface in the White Paper is the potential for dust to be  
16    generated by wind from treated fields. This is a remarkably  
17    little researched area, although I think it's potentially  
18    important. The limited data that are available in the published  
19    literature, I'm not aware of any unpublished registrant studies,  
20    indicate that in certain fine fractions of dust generated from  
21    fields, in particular something we define as PM10. Probably  
22    most of us in the room are familiar with that.

23          You can have very strong enrichment of  
24    pesticide active ingredients. So dust, i.e. generated from  
25    wind-blown materials from fields, can have relatively high  
26    pesticide concentrations. Therefore, I think we perhaps need to



1 step back and contemplate that a little bit and see if this may  
2 be an exposure pathway for bees, which would include a direct  
3 deposition of the dust on hives that are actually remote from  
4 the field as well as body contact from dust that accumulated on  
5 various surfaces, including plant surfaces.

6 I strongly feel that there are two parts of  
7 the dust question that need to be examined and relative to  
8 potential exposures in this case.

9 **DR. DANIEL SCHLENK:** Dr. Hunt:

10 **DR. GREG HUNT:** Well, I'll just speak to the  
11 seed treatment. Neonicotinoid seed treatments in particular, in  
12 the conceptual model, this is modeled - the EPA White Paper is  
13 only looking primarily at systemic movement plant parts. But  
14 clearly, we're seeing a problem with dust, particularly with  
15 corn planting and in regards to the soil. We see at least twice  
16 the concentrations of Clothianidin that we find in corn pollen.  
17 We're seeing a lot of reports, many of which apparently aren't  
18 getting transmitted to the EPA, and I think there is a lag in  
19 that also because, for example, in Indiana, the office of the  
20 state chemist has looked at 14 incident reports and they all  
21 came up positive for Clothianidin.

22 In Ohio, there was something like 50 reports,  
23 incident reports, which again have not gotten their way to the  
24 EPA. In Ontario and Quebec, there are a lot of positive reports  
25 - over 130 of them, I understand - just from this year. Last  
26 year in the midwest, there were no incident reports. The



1 difference is that now the beekeepers know about it, so they're  
2 making reports.

3 I think dust through talc and abrasion of seed  
4 is a mode of exposure that needs to be addressed. We need more  
5 data on residues and wax comb, which we have virtually nothing  
6 from yet, but we're working on that, and more pollen samples.  
7 So, I think I'll leave it at that.

8 **DR. DANIEL SCHLENK:** Okay. And Dr. Pettis?

9 **DR. JEFF PETTIS:** Just a few comments to add  
10 relative to dust, I agree that it's generated a lot of interest  
11 and public opinion of things this year. I don't think in  
12 general - there probably are two ways that dust could become an  
13 issue. One is the seed treatment and the other is just general  
14 dust from sprayed crops.

15 I do feel though, however, as far as looking  
16 at exposure levels that even with corn and seed treatment that  
17 the dust exposure is going to be a temporary effect, temporary  
18 and short-term effect whereas the translocation throughout the  
19 plant and expression in say, corn pollen, would be more of a  
20 chronic effect. So, I don't want to sight of that in focusing  
21 on the dust. I do think dust is a real issue that should maybe  
22 have a solid box rather than a dotted box. In terms of relative  
23 to dietary exposure, I think we just need to keep in mind that  
24 the expression of systemics in pollen and nectar are probably of  
25 more concern.

26 **DR. DANIEL SCHLENK:** Okay. Mr. Pistorius?



1                   **MR. JENS PISTORIUS:** Thank you. I think I  
2 have to have a longer comment on this. Looking at the question,  
3 I think there are two questions in which data should be a useful  
4 Tier I exposure method. The second part of the question is  
5 basically what data may be needed to address the exposure route  
6 quantitatively.

7                   From the research that we've done at the  
8 Governmental Institution in the last year, I think we can say,  
9 I've had the honor to make a presentation to EPA on the 27th of  
10 July last year. We presented a tip of the iceberg of all these  
11 data. We can say that dust is a relevant route, which has to be  
12 integrated into the risk assessment. Nevertheless, I think on  
13 Tier I, so that's the answer for the first part of the question,  
14 we don't need any more data. Although there are scientific  
15 question still open, for instance, that we have the indication  
16 that compared in gram a.i. per hectare for instance or the oral  
17 and contact toxicity compared with dust to spray. There is a  
18 higher problem, a higher toxicity of dust compared to sprays,  
19 also when we compare the effects of the sprays and dust, between  
20 a spray application and a dust application, looking at the same  
21 gram a.i. per hectare with higher effects for dust.

22                   Nevertheless, I think we have the option to  
23 say for Tier I screening exposure method, we can say that it is  
24 enough. If we look at the acute oral and contact data that we  
25 get with feeding bee nectar. So, we get an indication that, you  
26 know, we have highly toxic substances for instance. Then the



1 question is what is out exposure? We have done a lot - and I  
2 think I must apologize, we haven't published all the data yet,  
3 we are still working on the science. We've done a lot on  
4 generating data of seed treatment quality, what does it mean,  
5 how can you improve it, then machinery, what does it mean, how  
6 does the culmination of the two, considering that you can use  
7 deflectors with different efficacy, translate into potential  
8 exposure off crop.

9 With this off-crop exposure, we think is it  
10 really important issue that has to be considered. There would  
11 be data available that actually allow integration into the risk  
12 management process. What I believe is that the management  
13 issues that have been considered by EPA are an effective way of  
14 mitigating the risks if you have a quality assurance of the seed  
15 treatment quality and also the planting practices.

16 But in general for this answer, I can only  
17 give a limited answer for the second part of the question. The  
18 data that may be needed to address the exposure route  
19 quantitatively, I think that for highly toxic substances, you  
20 may have to go for higher tier tests.

21 **DR. DANIEL SCHLENK:** So, it sounds like a  
22 little bit of disagreement there. Some would say yes dust and  
23 Mr. Pistorius says no dust, I guess, in terms of the Tier I, is  
24 that what I hear?

25 **MR. JENS PISTORIUS:** Jens Pistorius. I think  
26 what you can do in the oral and contact laboratory tests, you



1 get an indication of how toxic is such a substance. If you have  
2 highly toxic substance, those are a matter of concern. We can  
3 clearly state from our findings that no toxic substances are not  
4 a matter of concern. Regarding the question - do we need an  
5 extra test to assess the toxicity of dust compared to liquid  
6 formulations in a laboratory level? No, we can extrapolate from  
7 the dust toxicity from the feeding toxicity and normal toxicity.

8 **DR. DANIEL SCHLENK:** I think this is just  
9 looking at exposure though, not necessarily toxicity. So, I  
10 guess the question - I could be wrong here, and other Panel  
11 members, but in terms of the Tier I exposure side is should dust  
12 be added into the other two variables. I guess that's how I see  
13 the question being asked.

14 **DR. THOMAS POTTER:** I say yes.

15 **DR. DANIEL SCHLENK:** That's what I thought.  
16 Yes?

17 **DR. JEFF PETTIS:** If I understood you  
18 correctly that the difference between the dust application and  
19 oral or dermal, you could gain the same information by.

20 **DR. DANIEL SCHLENK:** So the consensus is yes,  
21 or dust should be included.

22 **DR. THOMAS POTTER:** Can I get in here, because  
23 now I'm confused. This is Tom Potter here. When we're talking  
24 about dust, are we talking about dust that was generated from  
25 treated seed? Are we talking about dust as a formulation of the  
26 pesticide? Or are we talking about the dust associated with



1 wind-blown material from treated fields? So, I think we need to  
2 be clear in terms of what type of dust we are talking about in  
3 terms of exposure here. I'm a bit confused on that.

4 **MR. JENS PISTORIUS:** Jens Pistorius. Thank  
5 you for this because I forgot to mention that it's not only seed  
6 treatments, which may generate such problematic dust. Also  
7 granules may. What we did is we did investigations with abraded  
8 dust. So basically, it's the formulation.

9 **DR. DANIEL SCHLENK:** Yeah, Dr. Hunt?

10 **DR. GREG HUNT:** I think there is some  
11 consensus. I think we're talking about two different things in  
12 a way, but I think there is consensus among the group that  
13 abraded dust from treated seed should be considered on  
14 off-target plants as another method of exposure and that we need  
15 to - and I think that's clear. Just looking at the  
16 concentrations that we see in bee collected pollen, I think that  
17 as my colleague, Christian Krupke mentioned, that this is an  
18 area where clearly there could be some mitigation, planters  
19 could be modified. We could, as Tom Potter mentioned, estimate  
20 how much dust is coming out of the planter. There could be  
21 changes in formulation. There could be other regulatory actions  
22 or reanalysis of the regulation of this using so much  
23 neonicotinoids over such a large acreage, especially since it  
24 has such a long half life than high water solubility and such  
25 high toxicity.

26 **DR. DANIEL SCHLENK:** Okay. Any other



1 comments? Dr. Pettis?

2 **DR. JEFF PETTIS:** Well, I guess I would just  
3 like to hear maybe from the group or from EPA about EPA was  
4 proposing that mitigation measures might be the primary means of  
5 dealing with this dust issue. I don't know how the Panel feels  
6 about that, well other than - in other words, are mitigation  
7 measures at the time of planting sufficient for this issue?

8 **DR. DANIEL SCHLENK:** Okay.

9 **DR. THOMAS POTTER:** This is Tom Potter. Can I  
10 just comment then because I brought that up because honestly, I  
11 have no idea how effective the mitigation measures might be. I  
12 went and googled around it, but I didn't come up with anything.  
13 So, perhaps there's something marvelously effective?

14 **DR. DANIEL SCHLENK:** Okay. Just, let's do one  
15 thing at a time. Is that something the Agency wishes to answer  
16 or has an answer for?

17 **DR. STEVEN BRADBURY:** It's a challenging  
18 combination of entities that come to bear on reducing dust. The  
19 equipment manufacturers, the seed planters, and other  
20 manufacturers, the pneumatics - I'm not an engineer, I don't  
21 pretend to be one, but you all get that part. Then there's the  
22 pesticide itself and the product in the sticking agent that the  
23 registrant, but also the seed company are in play there. Then  
24 there's talc or graphite or other components that are mixed in  
25 with the seeds so they don't in the planter. The seed treatment  
26 is a little sticky because it wants to hold the fungicide and



1 insecticide on.

2           So without getting too much into statutory  
3 stuff, the FIFRA label only goes so far in terms of what we can  
4 or can't do in terms of asking for data. Then there are things  
5 we do that aren't on the label, but part of the stakeholders  
6 getting together and trying to work through some solutions.  
7 There was a public meeting mid July, end of July at Iowa State  
8 and the State of Iowa convened in sort of an open meeting to  
9 sort of brain storm on this very topic. Some companies were  
10 describing with how they're coming up with some alternatives to  
11 talc, and they're working with the seed planting companies and  
12 they're working with registrants and they're working with  
13 formulators. In fact, there is equipment that can be used to  
14 measure how much dust is coming off the benchtop in the  
15 laboratory, sort of reasonable surrogates to get a sense of  
16 different combinations of material can change the amount of  
17 abrasion that occurs, and you can measure the amount of the  
18 pesticide in that dust.

19           So, as far as data, it appears that there are  
20 people working on the kinds of experiments where you could start  
21 to get some insight, which then could give us some ways of  
22 between what you could potentially do. I'm not trying to  
23 project too far into the future, but theoretically some things  
24 you might be able to do on a FIFRA label versus things that play  
25 out in best management practices, things USDA and others could  
26 be doing in terms of improving DMPs, which may be related to



1 dust management in general. There is probably going to be a  
2 combination of data we can get and some experiments to get  
3 insights into how products are used.

4 **DR. DANIEL SCHLENK:** Okay. Does that answer  
5 your question Tom?

6 **DR. THOMAS POTTER:** Yes, I think very well.  
7 Obviously it is in our complex set of characters involved. If I  
8 could just offer one more comment on this, I think that by  
9 including some estimate of exposure associated with dust  
10 unmitigated, it may in some sense motivate, force, compel other  
11 parties to you know, increase their effort in this mitigation  
12 area.

13 **DR. STEVEN BRADBURY:** I would like to say one  
14 thing that information that was shared at this meeting at Iowa  
15 State wasn't the worst bias to the credit of the seed planting,  
16 the John Deere and the other companies and folks working on  
17 formulations and some registrants. They were working on it on  
18 their own. So, I think people are working hard and trying to  
19 figure out the engineering and the chemistry and the efficacy of  
20 the material for plant protection. And there were beekeepers at  
21 this meeting. It was quite a good meeting actually, people  
22 trying to come up with solutions.

23 **DR. DANIEL SCHLENK:** That was Dr. Bradbury. I  
24 think we had Dr. James first? You put your hand back down.

25 **DR. ROSALIND JAMES:** I just wanted to  
26 reiterate, I guess, about the dust from pesticide applications



1 that are not soil treatments also, and make sure that doesn't  
2 get lost in this discussion.

3 **DR. DANIEL SCHLENK:** And Dr. Berenbaum?

4 **DR. MAY BERENBAUM:** I just was not clear on  
5 whether EPA has regulatory enforcement authority over dust? I  
6 mean, is that within bailiwick or.

7 **DR. STEVEN BRADBURY:** Not under FIFRA. If  
8 there is a formulation that is a dust, then that is covered. If  
9 it gets a little less - if it's dust blowing off of soil from  
10 soil on a field that has been in corn for two to three to four  
11 years, and there is probably something in there along with the  
12 nitrogen and phosphorus. That blowing around - FIFRA is not  
13 capturing that. It could be aspects of the Clean Air Act that  
14 are capturing that. You have probably heard of the PM10 rule  
15 and there is a PM10 rule now in the agricultural setting that is  
16 working through. So, without projecting too far, there is  
17 probably some air mottling that Office of Air did try to  
18 understand PM10 movement. There may be a way to tap into some  
19 of that at the bounding estimates, but you start to get the  
20 shade of gray in terms of where one statute drops off and  
21 another one starts.

22 **DR. DANIEL SCHLENK:** That was Steve Bradbury.  
23 Dr. Hunt?

24 **DR. GREG HUNT:** I guess I'm a little out of my  
25 element here because I'm not a toxicologist. I'm just a bee  
26 researcher, but I guess we're just looking at the risk



1 assessment side. We're not looking at the benefit side. To me,  
2 that is an unknown because we don't have access to these yield  
3 studies. I don't know if they've been done just with  
4 neonicotinoids or with the entire seed treatment mixture, but it  
5 seems like this already registered product is being used on a  
6 wide scale. It's kinda of an experiment ongoing. I've talked  
7 to the chemical companies. They are working on litigation and  
8 they've shown me data, how different seed treatments come off  
9 the seeds more than others. Corn is the worst in that regard.  
10 I guess I'm wondering what is the EPA thinking in the big  
11 picture with neonicotinoid seed treatments. I don't know about  
12 regulatory laws very much. Is there any consideration.

13 **DR. DANIEL SCHLENK:** Dr. Hunt, that's getting  
14 a little bit into policy and sort of away from the question that  
15 we're being asked at this particular point. Let me try to bring  
16 us back to that question, which is basically should dust be  
17 included in the Tier I assessment and what other ways do we  
18 quantify that. From what I understand is that we are in  
19 consensus that dust should be used. Apparently, there have been  
20 several recommendations of different methods to do that. Is  
21 there anything else that we need to add on that? Yes, Mr.  
22 Pistorius?

23 **MR. JENS PISTORIUS:** Jens Pistorius. I think  
24 dust is not only an issue for maize, maybe also for (inaudible),  
25 but there is large number of other crops where dust is not an  
26 issue at all because of the seed treatment quality, which is



1 very different for different seeds.

2 **DR. DANIEL SCHLENK:** If you can, again, give  
3 those comments to Dr. Potter to simulate that information, that  
4 would be great. Dr. Hunt?

5 **DR. GREG HUNT:** I'm sorry, but the question  
6 actually was should dust be considered as a route of exposure,  
7 and I think we addressed that.

8 **DR. DANIEL SCHLENK:** Yes, that's what I said,  
9 yep. Okay. Any other comments on letter B? Okay. Let me go  
10 to Ms. Wendel. Are you clear with Panel's answer? Question or  
11 clarification?

12 **MR. KEITH SAPPINGTON:** Clarifying question -  
13 Keith Sappington. If we would be interested in the methods of  
14 how you would incorporate a dust estimate of exposure a priority  
15 in the Tier I assessment.

16 **DR. DANIEL SCHLENK:** Okay. I think we are  
17 clear on that. Okay. Let's move onto C, and read that into the  
18 record please.

19 **MS. CHRISTINA WENDEL:** Christina Wendel,  
20 question number 7, subpart (c), drinking water -- please comment  
21 on the analysis, discussions provided in Appendix 2 of the White  
22 Paper and the conclusion that pesticide exposure to bees through  
23 drinking contaminated water is not expected to be a major route  
24 of exposure when compared to contact following foliar spray  
25 applications and diet. If the Panel believes that this exposure  
26 route should be quantitatively included in the Tier I exposure



1 method, for the relevant application types, i.e. foliar spray,  
2 soil treatment, seed treatment, or trunk spray, please discuss  
3 why and what data may be needed to address the exposure route  
4 quantitatively.

5 **DR. DANIEL SCHLENK:** And Dr. Potter.

6 **DR. THOMAS POTTER:** Okay. Well, I think it  
7 was said earlier that we felt like it was important for this  
8 route of exposure to be included. I believe that Tier I, we  
9 haven't had a discussion among the group here. So, we may  
10 disagree, but I don't think we will. So, the short answer is  
11 yes, it should be included.

12 Okay. Now the second part of the answer,  
13 we're going to need to look at two different issues that  
14 ultimately translate to exposure. One is potential  
15 concentrations of pesticides in water that bees may be exposed  
16 to. That was addressed to some degree in the appendix, and  
17 we'll get to that, but I just wanted to mention the second part,  
18 and that is the question of just how much water do bees and/or  
19 other pollinating organisms consume on a daily basis. Clearly,  
20 we need to multiply the two amount times concentration to come  
21 up with exposure. So, we will break out the answer into those  
22 two parts in terms of the comment.

23 First with regard to potential concentrations  
24 of active ingredients in water sources that bees may use, I  
25 believe and I think there will be consensus on that, that the  
26 concentration levels that we generally find in surface waters in



1     aquacultural and in urban areas across the United States are  
2     generally very low. Typical datasets that are available were in  
3     the parts per billion or subparts per billion level for most  
4     active ingredients. So, these are exceedingly low relative to  
5     potential exposures given estimated volumes of water consumed  
6     during the day. I would say, generally data that is out there  
7     supports the conclusion that water should not be concluded. So,  
8     there, I've backed up and gone in the opposite direction here,  
9     but we'll get there.

10           The second part of this is that the Agency in  
11     their appendix went through and did a couple of assessment of  
12     potential exposures. One of them, they used the model GENEEC,  
13     which is the standard model to use, screening model to be used  
14     to estimate pesticide exposures in drinking water and surface  
15     water. It's a first tier model. I believe that model is used  
16     correctly. Basically, it shows that even in a worst case  
17     scenario, if you are looking at a farm pond, which has a lot of  
18     water in it, obviously - I mean, I'm not saying that  
19     facetiously. So, there is a potential for substantial dilution.

20     The concentrations again are relatively low. So, that's not  
21     likely, again, to be a significant exposure pathway.

22           Finally, the third area that we may want to  
23     consider in this, is the idea of puddles that are left in the  
24     field, either from irrigation water or rain. Those can be  
25     directly sprayed as noted in the appendix. In their  
26     deliberations, Agency staff went through and did some



1 calculations using a simple equilibrium model that I think was  
2 extracted from a rice paddy assessment that was done regarding  
3 pesticides the use in rice paddies. I went in and I looked at  
4 that. I think that there was an error in the calculations. I  
5 will provide this in written comment. That error resulted in  
6 the estimated concentrations in puddles being essentially 1000  
7 times lower than what they would likely be using the same set of  
8 assumptions. So, anyway, I think that ends up indicating that  
9 puddles that are in fields that are sprayed may have relatively  
10 high concentrations of pesticides. We're talking about in the  
11 parts per million level. If we use those values and then go  
12 into looking at the volume of water that bees may consume on a  
13 daily basis.

14 First, if we use the low value, which was  
15 recommended, I think, from the paper wasp study -- is that  
16 correct? Forty-five microliters a day, if we multiply that  
17 times these estimated high-end estimated concentrations in  
18 pesticide contaminated puddles, the drinking water exposure  
19 could be somewhere in the order of 10 percent to 20 percent of  
20 the dietary exposure. So, I think that's a potentially  
21 significant exposure pathway and should be included.

22 Further, if we go to the second part of the  
23 exposure equation, which is that the volume of water consumed, I  
24 certainly would like to hear from some of the other panel  
25 members who are more knowledgeable about bees than I am. It  
26 struck me that there was a substantial uncertainty with regard



1 to the volumes that may be consumed on a daily basis. The  
2 assessments that were made indicated they varied from somewhere  
3 in the order of 45 microliters a day to 6 milliliters a day,  
4 which is a factor of about 100.

5 So, if we do some bounding estimates here  
6 using those high-end concentrations that we could estimate by  
7 doing our equilibrium model for pesticide-contaminated puddles.  
8 Drinking water exposure could indeed exceed dietary exposure.  
9 So therefore, I strongly feel that it needs to be captured in a  
10 Tier I assessment, and perhaps some effort could also be made in  
11 terms of further refining potential environmental  
12 concentrations. I made this comment yesterday in the form of a  
13 question. But in ECOFRAM, there is a module that actually  
14 looked at runoff in fields and accumulation in puddles using  
15 some subcomponents of PRZM, relatively simple model to use and  
16 run. Perhaps that should be an alternate approach in terms of  
17 estimating pesticide contamination in field puddles that might  
18 provide some insight into this exposure issue.

19 **DR. DANIEL SCHLENK:** Thank you, Dr. Potter.  
20 Dr. Hunt?

21 **DR. GREG HUNT:** I'm not sure, but I believe  
22 that bees get most of the dietary water through nectar. I would  
23 need to look in the literature to confirm this. They will  
24 collect water in the fields or in puddles primarily to cool the  
25 hive, the deposit in the comb, to cool the hive and also to  
26 dilute the brood food, but they will also use nectar for that.



1 That's my understanding.

2 **DR. DANIEL SCHLENK:** So, I guess you would say  
3 the water should not be included - is that.

4 **DR. GREG HUNT:** I'm saying I don't know.

5 **DR. DANIEL SCHLENK:** Okay.

6 **DR. THOMAS POTTER:** Can I ask a followup on  
7 that?

8 **DR. DANIEL SCHLENK:** Sure, yeah.

9 **DR. THOMAS POTTER:** This is Tom Potter here.  
10 I think what we're talking about is around the lunch break, and  
11 I was asking how do the bees get the water back to the hive?  
12 What my understanding was that they were essentially ingesting  
13 it and then regurgitating it.

14 **DR. GREG HUNT:** Yes, that's right. They  
15 ingest it into the honey stomach, which is like a waterproof bag  
16 more or less, but they have the pyloric valve from the honey  
17 stomach to the midgut and some water could pass through that.  
18 And I suppose pesticides could be absorbed through the walls of  
19 the stomach.

20 **DR. DANIEL SCHLENK:** Okay. Dr. Pettis?

21 **DR. JEFF PETTIS:** I'll just a bit to what Dr.  
22 Hunt said about water used by bees. Primarily for two reasons,  
23 as you said. One is to dilute honey to then use as food and  
24 also to dilut brood food somewhat. The big issue - so the most  
25 volume is in cooling the hive. There again, certain bees go  
26 out, they collect the water and bring it back in. They can



1 actually sit in the hive and hold his water in their body for  
2 extended periods of time, act like little water tanks held  
3 within. Then, they move it around the hive and evaporate it so  
4 it can be put into cells and evaporate in the cells. So, at  
5 certain times, and in climatic conditions where you have hot  
6 weather, the use of water can be extensive.

7 I would probably agree with you Greg, that  
8 that in general under non-hot conditions that there is a lot of  
9 - the water requirements are probably met by consuming honey  
10 and/or nectar. But when you have hot conditions, certainly the  
11 increase for cooling needs is great. So, I think from my  
12 perspective, I would think that it can't be discounted as a  
13 source of exposure. How will you incorporate it into the Tier I  
14 - I'm uncertain at this time.

15 **DR. DANIEL SCHLENK:** Mr. Pistorius and then  
16 Dr. Fefferman?

17 **MR. JENS PISTORIUS:** I have a proposal how to  
18 integrate it in Tier I risk assessment, and it will be published  
19 in about one or two weeks along with the run of data on  
20 guttation issues published in the proceedings of the ICPBR.  
21 Basically, what we've done in Germany again, is a lot of  
22 different crops. We looked at the residue that get into the  
23 guttation droplets and then also, we did semi-field and field  
24 trials with worst case and realistic exposure. What you can say  
25 - okay in semi-field, you get high mortality if you don't give  
26 any other water to the bees, but if you give another water



1 source, it is quite lower.

2 Now to the field test and monitoring that we  
3 have done, we have had, not only by us but agricultural estates  
4 and institutes - about 12 different monitoring with a lot of bee  
5 hives and worst case exposure directly at maze fields.

6 In one of the trials, we had on two days  
7 increased mortality. So, as a general answer to the question  
8 that is asked here - what was it? Drinking guttation water is,  
9 in my opinion, not expected to be a major route of exposure when  
10 compared to contact and diet is what I agree upon. I'm not  
11 quite sure what it is like here for all the desert situations.  
12 That is maybe different. I can only say from my findings. What  
13 we can do is have a little bit of distance, so it is a  
14 mitigation measure between highest and the crop, whoever has to  
15 keep this distance.

16 **DR. DANIEL SCHLENK:** Dr. Fefferman?

17 **DR. NINA FEFFERMAN:** So, while I don't know to  
18 speak to the drinking water per se, I agree that most of the  
19 water collected is not being used for drinking. I think that  
20 same intent - although maybe this comes under other exposure  
21 routes and I don't know, its still water. By virtue of the fact  
22 that that water is being collected for cooling, I think plays  
23 directly into what we were talking about with the answer to the  
24 early question about wax exposure.

25 As it's circulated around the wax and  
26 evaporated, I don't know how much pesticide evaporates with that



1 water and how much is left residually on the wax. I think that  
2 actually might be, especially at the hottest points in the  
3 summer, a substantial introduction of pesticide into the wax  
4 comb itself and that may be a temporal aspect of all of this  
5 that might be important to look into.

6 **DR. DANIEL SCHLENK:** It sounds like a seasonal  
7 component for sure. Any other comments? Yep, Dr. Tarpay?

8 **DR. DAVID TARPAY:** I just wanted to reiterate  
9 that point too, is that if using mostly for cooling, the three  
10 studies that are cited in Appendix 2, at least one of them is  
11 really done in upstate New York, right? Where it doesn't get  
12 above 70 degrees in July, right? So, in other places, Central  
13 Valley of California, with 120 degrees and the bees keeping the  
14 brood nests at 95 degrees, it takes a substantial amount of  
15 water for that cooling to occur. So, I would say that those  
16 estimates in worst case scenarios could be incredibly low, but  
17 there seems to be a data gap there obviously and very high water  
18 needs, not drinking water, but high-cooling water needs.

19 **DR. DANIEL SCHLENK:** Dr. Berenbaum?

20 **DR. MAY BERENBAUM:** Shame on me for  
21 introducing anecdotal unscientific observations here. I'm  
22 asking my bee keeping colony - I'm not a bee biologist, but I'm  
23 the one who gets the calls from people who want to save the bees  
24 and help the bees. I can't tell you how many calls I've gotten  
25 from people who have had bees that drown in bird water baths -  
26 that's the word. So, my impression is that a tiny bit



1 unscientific, I can't site literature here, but bees do visit  
2 water and get immersed in water. So, that's totally anecdotal,  
3 but that may be something to consider, particularly because  
4 there can be puddles that vary in their pesticide content.  
5 That's not unexpected. I don't know how that fits into  
6 calculations of exposure.

7 **DR. DANIEL SCHLENK:** Yes, Dr. Pettis?

8 **DR. JEFF PETTIS:** We would agree with you that  
9 bees do visit water and they need water. We were asked to  
10 comment on the exposure method relevant to the application  
11 types, foliar spray, soil treatments, seed treatments, trunk  
12 spray. There might be an important component here for the type  
13 of chemical involved, whether it is water soluble or insoluble  
14 in water. I would think that maybe the agency could take into  
15 account - if it's insoluble in water, then only foliar  
16 applications may be an issue were it collects in puddles or  
17 temporary things, whereas water soluble compounds, soil  
18 treatments, seed treatments, et cetera, could move through the  
19 soil and accumulate in water. So, there could be, based on the  
20 chemical type, we may consider that. Some of the studies  
21 looking at residues in standing water or temporary water,  
22 they've looked at Imidacloprid, which is water soluble.

23 **DR. DANIEL SCHLENK:** Yes, Mr. Pistorius?

24 **MR. JENS PISTORIUS:** I completely agree. It's  
25 no problem for a nonsystemic and only for highly toxic - it may  
26 only be a potential problem for highly toxic substances.



1                   **DR. DANIEL SCHLENK:** Okay. Anyone else?

2       Sounds like you got a range of answers there. Sounds like  
3       definitely maybe on that one. So, we will leave that to Dr.  
4       Potter to put all of those answers together in some coherent  
5       fashion there. You've heard several answers here. Do you have  
6       any questions or clarification?

7                   **MR. KEITH SAPPINGTON:** Keith Sappington.

8       Yeah, just methods for quantifying it, but yes, drinking maybe.  
9

10                  **DR. DANIEL SCHLENK:** Okay. Is that clear to  
11       you?

12                  **DR. THOMAS POTTER:** Tom Potter here. I just  
13       want to say, you know, and thanks for all the discussion here, I  
14       think this has been very informative, certainly to me, but it's  
15       not just drinking water. Obviously, it is water. And so, we  
16       ought to refine our terminology here to say water used.

17                  **DR. DANIEL SCHLENK:** Sure. Good point. Okay.

18       Let's move onto - I think this might be our last one of the day  
19       here, perhaps, letter D.

20                  **MS. CHRISTINA WENDEL:** Christina Wendel.

21       Question 7, subpart (d), other routes - please identify and  
22       discuss additional exposure routes, if there are any besides  
23       contact with dust and consumption of drinking water, that would  
24       contribute significantly to pesticide exposure of bees and  
25       explain how and why such exposures could be considered  
26       quantitatively in establishing the Tier I exposure value.



1                   **DR. DANIEL SCHLENK:** Dr. Potter?

2                   **DR. THOMAS POTTER:** Well, I think I'm going to  
3 send this over the Rosalind - just teasing, but hopefully you'll  
4 comment on this. I feel that the route of exposure that would  
5 need to be considered here would be direct soil contact. I  
6 think Rosalind described earlier that there are many bees who  
7 come in directly in contact with soil using the material for  
8 nesting, nest building and et cetera. So certainly, there is  
9 potential for contact. Now the question of how we would  
10 evaluate that quantitatively, I'm hoping somebody here is going  
11 to help me with that.

12                   **DR. DANIEL SCHLENK:** Dr. Hunt, any help for  
13 Dr. Potter there?

14                   **DR. GREG HUNT:** Not really. One other obvious  
15 route of exposure that we haven't talked a lot about is the wax  
16 comb itself with the brood swimming in royal jelly, which is  
17 liquifilic and the wax comb, which is liquifilic, but that would  
18 come under food, I mean ingestion or absorption through the  
19 cuticle. But, I don't think that needs to be, in my opinion,  
20 considered under Tier I exposure, but it's just another route of  
21 exposure.

22                   **DR. DANIEL SCHLENK:** So, not a significant  
23 route of exposure, not to be included in that first tier  
24 approach. Okay. And Dr. Pettis?

25                   **DR. JEFF PETTIS:** Really, not much to add that  
26 hasn't been already said, other than the fact that bees are



1 great samplers of the environment, and they have these hairs  
2 that will pick up all kinds of dust and particles and whatever,  
3 but it's not really - it's more of a general issue with bees,  
4 not so much a regulatory issue.

5 **DR. DANIEL SCHLENK:** Well, what do you say,  
6 Dr. James - are you going to help Dr. Potter out there?

7 **DR. ROSALIND JAMES:** I'm probably just  
8 reiterated what I have said earlier today in that if you are  
9 going to use honey bees as a surrogate for other bees, you need  
10 to think about where other bees go besides just where honey bees  
11 go, and you should add contact toxicity for soil pesticides.  
12 So, you already have some estimates and some methods for  
13 estimating what concentration of pesticides should be in the  
14 soil, and I recommend adding larval contact toxicity test for  
15 pesticides that are in the soil, and the reasoning being you  
16 have your soil nesting bees and you also have mason bees that  
17 are using mud for agricultural fields.

18 **DR. DANIEL SCHLENK:** Okay. Anyone else? Oh  
19 sorry, Dr. Hunt?

20 **DR. GREG HUNT:** Just a question for Rosalind.  
21 So, that would be kind of like the brood food. I mean, if you  
22 spike the brood food, then there are contacts with that, right?  
23 Would that cover it?

24 **DR. ROSALIND JAMES:** But this has to do with  
25 exposures and your estimating exposure levels from soil, I guess  
26 is the difference. What is a contact exposure level from soil?



1  
2 **DR. DANIEL SCHLENK:** Okay. Any other comments  
3 for question 7? Going once, going twice. Okay. Let me go to  
4 the Agency. Are you clear or have any questions or  
5 clarification in terms of the Panel's response? We're good to  
6 go. Okay. Well, let's call it here for a break until tomorrow.  
7 We've made great headway. Congratulations to the Panel and  
8 appreciated for your hard work and meeting together in getting  
9 these things efficiently done without redundancy. We are about  
10 one-third of the way done, a bit over a third of the way in  
11 terms of number of questions, although there are 14 total  
12 questions - there's seven, but there are a lot of other subparts  
13 that we have to go through yet. So with that, I will turn it  
14 over to Fred for some administrative comments.

15 **DR. FRED JENKINS:** I don't have a whole lot  
16 extra to add, but I just want to thank the panel for a good day  
17 of deliberations and also the public commenters. I think we  
18 accomplished a lot today. So, I just wanted to say to the  
19 panel, remind them to please return your thumb drives to us with  
20 the presentation, so we can have those recycled. And with that  
21 said, we will see everyone in the morning at 9 a.m. Have a  
22 great evening.

23 (WHEREUPON the meeting was adjourned for the  
24 day.)

25 **DR. DANIEL SCHLENK:** Good morning everyone. I  
26 think in lieu of time, we are going to sort of skip our



1 introductions since, I think everybody know everyone by now, I  
2 hope. This morning, the Agency is going to hit us with some  
3 questions of clarification, I believe, on question 7, is that  
4 correct? Yep, anybody who is going to be doing that. Okay.

5 **MR. KEITH SAPPINGTON:** Thank you, Dr. Schlenk.

6 I would like to offer, on behalf of the Agency, just a little  
7 question of clarification on charge question 7, but first, we  
8 want to thank again the Panel for all their work over in  
9 preparing for this meeting and the deliberations of the last  
10 couple of days. We've already gained quite a lot of insight on  
11 how we can improve the proposed process. So, we really  
12 appreciate that.

13 With regards to question 7, and I know the  
14 Panel has all ready considered this, so this is perhaps more of  
15 an emphasis and that is in considering the alternate pathways in  
16 Tier I exposure, it would be helpful to us in understanding the  
17 conditions in which considering these additional pathways, like  
18 drinking water and dust in associated oral exposure from dust  
19 onto other plants, conditions in which that might change the  
20 outcome of the Tier I assessment from say passing the assessment  
21 to not passing the assessment since our goal is to minimize type  
22 2 errors.

23 So, in thinking about that and then in  
24 thinking about the different context, so perhaps for example,  
25 for the seed treatment where we have a default value of 1 mg per  
26 kilogram. Is there is enough margin and safety such that not



1 including drinking water or some of these other sources, would  
2 that likely really push that from a no risk or no LOC exceedance  
3 to a yes exceedance. Thank you very much.

4 **DR. DANIEL SCHLENK:** I believe that Dr. Potter  
5 is the lead on question 7, so do you have any questions related  
6 to how you want to present that?

7 **DR. THOMAS POTTER:** Well, I have a comment. I  
8 probably have to come up with a few questions too here. Anyway,  
9 good morning, Tom Potter here. Are you specifically asking  
10 about the seed treatment scenario only or all of the scenarios  
11 that we were looking at?

12 **MR. KEITH SAPPINGTON:** Keith Sappington. All  
13 the scenarios and considering the alternate routes.

14 **DR. THOMAS POTTER:** Right. You know, one of  
15 the things that, you know, in terms of, let's just break them  
16 out here. I'll allow for a few thoughts on it. I saw Dr. Hunt  
17 also had a comment that he wanted to make, but you know one of  
18 the things about the dust is that hopefully there will be some  
19 effort to look at the potential exposures. Perhaps you've done  
20 that all ready. So, if indeed you could come to some well  
21 established conclusion that that is not an issue, it certainly  
22 may not belong in Tier I. This is my opinion.

23 So there needs to be some assessment,  
24 particularly in regard to wind-blow soil dust. That's kind of  
25 my issues. We will let Jens talk about the abraded material  
26 from seed coat in a few minutes perhaps.



1                   With regard to drinking water, I would offer  
2     the same line of thought or reasoning. I think that, you know,  
3     a reassessment to determine if there are potentially significant  
4     exposures. I think we noted yesterday that there was  
5     considerable uncertainty in the amount of water that bees drink  
6     and come in contact with on a daily basis, varying on a factor  
7     of a range of 100. So, certainly that could have a major impact  
8     on potential exposures.

9                   And then there was the question of assessing  
10    potential concentrations, in again a worst case scenario and  
11    overspray on a field that had standing water in it, using the  
12    type of exposure scenario to come up and say well, you know is  
13    this 1 percent of the dietary contact exposure or is it 50  
14    percent or is it greater than the dietary - I mean if you use  
15    the higher numbers for the water exposure in high end numbers  
16    for field puddles, you can come up with a pretty significant  
17    load. No, I don't necessarily think that that's reasonable or  
18    realistic thing to do, but I think it's something that you  
19    should spend some more time working on in the process of  
20    refining the White Paper.

21                   **DR. DANIEL SCHLENK:** Dr. Hunt?

22                   **DR. GREGORY HUNT:** Well, in considering, for  
23    example, the abraded seed coat dust, I don't think that  
24    considering that route of exposure would affect your Tier I, you  
25    know, because 1 mg per kilogram is very conservative. It might  
26    affect the downstream, for example, and I look at the figures on



1 the tiered assessment. Downstream if it exceeds the LOC, then  
2 it says use available crop residue studies, recalculate our  
3 cues. So, in that blight, the crop residue studies may be  
4 should include like incoming pollen that the bees are bringing  
5 in. What are the maximal values you are seeing in those?

6 **DR. DANIEL SCHLENK:** Dr. Pettis, you're also  
7 on this one too.

8 **DR. JEFF PETTIS:** I have no immediate comments  
9 at this time.

10 **DR. DANIEL SCHLENK:** Dr. Fefferman, and then.

11 **DR. NINA FEFFERMAN:** Thanks. I think one  
12 major thing I took away from the conversation yesterday from 7  
13 is maybe even the concept of the phrase drinking water might be  
14 an inappropriate way to frame that, and that just water usage  
15 might be a way to frame that. While I don't know, again,  
16 whether or not on testing this will turn out to be significant.  
17 It does lead, in my mind, to the suggested additional outcome  
18 that maybe wax sampling concentrations should be measured as  
19 part of a Tier I assessment, because that could tell us a  
20 gradual accumulation over time as opposed to a dose response  
21 from an initial carrying of what exposed larvae are facing or  
22 what recycling of wax within the hive is doing.

23 So, if we're worried about deposition  
24 gradually over time from evaporation leaving not so much  
25 drinking water but evaporated water for cooling leaving residues  
26 of pesticides within the wax. Then that suggests to me not only



1 a potential for it to trigger a positive case where maybe it  
2 should be. Not only false but true, but also a new mechanism to  
3 measure, which is the concentration in the wax.

4 **DR. DANIEL SCHLENK:** Okay. Mr. Pistorius?

5 **MR. JENS PISTORIUS:** I want to thank EPA for  
6 this excellent question. No, it will not change the outcome of  
7 the risk assessment and the Tier I seems to be conservatively  
8 enough that the substances of concern are identified. So, it is  
9 an issue of higher tier testing to address if there are concerns  
10 under realistic conditions, but it will not change. Also the  
11 route of dust and guttation, and I think also for the risk of  
12 water, a lot changed the outcome of the risk assessment. You  
13 will be able to identify the substances of concern in Tier I.

14 **DR. DANIEL SCHLENK:** Okay. And Dr. James, did  
15 you have your.

16 **DR. ROSALIND JAMES:** Wouldn't it change a risk  
17 assessment for nonsystemic pesticides applied to the soil with  
18 the dust because the dust would provide another route of  
19 possible exposure that you didn't account for with a  
20 nonsystemic? You talk a lot about nonsystemic soil-applied  
21 pesticides.

22 **DR. DANIEL SCHLENK:** Mr. Pistorius?

23 **MR. JENS PISTORIUS:** I think it is both for  
24 the systemic and nonsystemic that the Tier I level will be able  
25 to identify those substances of concern where you find a high  
26 toxicity and you have a classification as highly toxic



1 substance, which will detect in the laboratory, oral and contact  
2 toxicity tests. You will be able to identify substances with  
3 potential concern. For nonsystemic substances, guttation is not  
4 an issue at all, but I agree dust is for highly toxic  
5 substances.

6 **DR. ROSALIND JAMES:** Rosalind James. But I'm  
7 out for dust, not guttation. Right.

8 **MR. JENS PISTORIUS:** Yes, as I said for dust,  
9 highly toxic substances are a concern no matter if systemic or  
10 nonsystemic.

11 **DR. DANIEL SCHLENK:** Anyone else want to weigh  
12 in on this one? Okay. Mr. Sappington, does that answer  
13 questions? Are you guys good with that?

14 **MR. KEITH SAPPINGTON:** Thank you very much.  
15 It helps a lot.

16 **DR. DANIEL SCHLENK:** Okay. Great. Okay. At  
17 this point in time, I guess we are ready to move onto question  
18 8, and if you could read in letter A for question 8?

19 **MR. JOSEPH DECANT:** Good morning. This is Joe  
20 Decant of EFED. So, I'll start us off on the toxicity side.  
21 Question 8, subpart (a) - please comment on the extent to which  
22 currently available bee toxicity tests, which focus primarily on  
23 mortality or survival, serve as an effective Tier I screen.

24 **DR. DANIEL SCHLENK:** And our lead discussant  
25 is David Tarpy.

26 **DR. DAVID TARPY:** Thank you, Mr. Chairman.



1 Good morning everybody. This is David Tarpy with North Carolina  
2 State University. I just want to thank the EPA and those who  
3 crafted the White Paper are really doing an excellent job. It's  
4 been incredibly informative to me. It's been obviously very  
5 thorough, very well articulated, and I think very helpful to  
6 have this discussion. I very much welcome it and I'm thrilled  
7 we're all doing it.

8 I foresee our conversations today looking at  
9 the effects on honey bees as a proxy for all bees and  
10 pollinators as being very important because the use of a social  
11 system to study solitary systems in general, it adds many  
12 complexities of biological organization that need to be taken  
13 into account, particularly at Tier I types of bioassays. So, I  
14 think we have a lot of thoughts concerning that that we'll be  
15 discussing today.

16 A couple of points that the discussants for  
17 this particular question wanted to bring up before we get into  
18 other details and later sub-questions and later question, one is  
19 to reiterate the issue of using the individual bee as the basal  
20 determinator rather than standardizing for body weight of the  
21 individual, especially if honey bees are going to be used as a  
22 proxy for other types of bees that can vary quite dramatically  
23 in their size. Seemingly to standardize on a milligram per bee  
24 is going to be much more translatable to those other symptoms  
25 than just doing it on a per bee basis.

26 Also, because honey bees are obligatorily



1 social as the White Paper very well articulates, they're a super  
2 organism, and that introduces a lot of important biological  
3 phenomenon to measuring them in vitro and in vivo. Sometimes  
4 those can be tested - sometimes honey bees can be tested in a  
5 vacuum as an individual, but often times, it's not very  
6 biologically meaningful. So, including the social context in  
7 Tier I screenings I think is going to become very important.

8 Another aspect, and this derives from the  
9 social complexity of honey bees as social insects is that a lot  
10 the Tier I screening techniques that's commonly used for  
11 solitary insects don't really capture a temporal component to  
12 the potential effects of screening for these compounds. Cutting  
13 it off at that 48 hours or extending that to 72 or even 96 hours  
14 doesn't really capture a lot of the social effects over time.  
15 So, adding that additional dimension to these bioassays we think  
16 would be very informative, and we'll be discussing those in a  
17 little more detail.

18 Specifically to address the need for measuring  
19 outcomes of individual bees in a social context, luckily there  
20 are bioassays that are routinely used in apicultural and honey  
21 bee biology research that can do this in vitro very high  
22 throughput and very readily, very reliable. For example, Evans  
23 et al. 2009 published a paper that just articulates the use of  
24 what they call bee cups, which are placing dozens, usually  
25 around 50 or so newly emerged adult honey bee workers into  
26 plastic cups, clear plastic cups where they are fed at lib with



1 food and can be subject to various treatments, and then in an  
2 incubator followed over a period of time.

3 So, it seems logical to the Panel who weighed  
4 in on this that there can be two bioassays whereby that type of  
5 system could capture these types of phenomenon. One is - not to  
6 use acute versus chronic here, but rather kind of a one-time  
7 application of a compound in question and the following a marked  
8 cohort of individuals within that social group, within that very  
9 small social unit over time, and then looking at their  
10 longevity. So, measuring longevity curves within that group of  
11 a one-time application of a particular compound.

12 Another analogous means of doing that would be  
13 to feed the entire group consistently with different compounds,  
14 so that the entire group is constantly exposed to that and the  
15 following the same longevity over time to see how long the focal  
16 workers live. We can provide additional detail about the  
17 logistics of these bioassays, but I think the endpoint here is  
18 that studying them in a social context when it deals with honey  
19 bees I think is very important compared to isolating them and  
20 testing them in a vacuum, which has a lot less biological  
21 meaning to honey bees as a social system than doing it in a  
22 social context.

23 One last point to point out again to deal with  
24 his particular question about just looking at the currently  
25 available toxicity test with looking at mortality and survival  
26 as endpoints is also another temporal component and that is the



1 seasonality of honey bees themselves and how honey bees can be  
2 physiologically different over different times of the year.  
3 I'll let Dr. Ostiguy comment on this further if she wants, but  
4 bees in the summer are very physiologically different from bees  
5 in the winter. So, incorporating the types of bees that would  
6 actually be used in these bioassays is biologically meaningful  
7 and nontrivial and out to taken into consideration on this.

8 **DR. DANIEL SCHLENK:** Dr. Berenbaum?

9 **DR. MAY BERENBAUM:** Fine. The only other  
10 thing - no I just see Ostiguy before Berenbaum. The only other  
11 - I concur, would just add to that there be useful to be at  
12 least cognisant of the genetic differences among populations and  
13 races, which is a well-known biologic attribute of bees among  
14 bee keepers. Dog breeds differ in the susceptibility to drugs  
15 or any issues with races and population.

16 **DR. DANIEL SCHLENK:** My apologies. Didn't  
17 mean to jump the gun there. My version is different than yours  
18 I think. Dr. Ostiguy?

19 **DR. NANCY OSTIGUY:** I concur with what David  
20 said. One thing about the summer/winter bees that we should  
21 also be taken into consideration when designing Tier I studies  
22 is that the particular exposures that you might see are  
23 different. So for instance, exposure to honey dew is going to  
24 be a fall phenomenon. Those will only be winter bees then. So,  
25 you can actually very much target where you think an exposure  
26 might be.



1 **DR. DANIEL SCHLENK:** Okay. Mr. Pistorius?

2 **MR. JENS PISTORIUS:** For the available bee  
3 toxicity tests as OECD 213 to 214, I think it is important to  
4 take into account behavioral abnormalities that are observed in  
5 the laboratory tests, especially if they occur at several doses  
6 that are clearly below mortality in a larger number of bees.

7 **DR. DANIEL SCHLENK:** Anyone else on the Panel  
8 want to get in on this one? Okay. Let me go to the Agency.  
9 Mr. DeCant, any questions of clarification? Okay. Let's move  
10 onto the second section then.

11 **MR. JOSEPH DECANT:** Joe DeCant, EFED.  
12 Question 8, section B -- please comment on additional  
13 measurement endpoints such as growth, which should be considered  
14 in future modifications of Tier I test protocols, and which are  
15 appropriately linked to the proposed assessment endpoints. Given  
16 that the queen is the reproductive unit of the colony, please  
17 comment on methods to evaluate effects on individual queens,  
18 considering practical limitations of testing with queens.

19 **DR. DANIEL SCHLENK:** Dr. Tarpy?

20 **DR. DAVID TARPY:** This is Dave Tarpy again, NC  
21 State. So this is where some of those other effects that Mr.  
22 Pistorius was just addressing, which are kind of the endpoints  
23 that we didn't articulate in part A. There are many - I think  
24 because of the social complexity of doing these bioassays in  
25 groups, it actually adds a lot of different phenomenon that can  
26 be measured and easily quantified and adds some robustness to



1 these types of bioassays. One case in point including the  
2 additional temporal component rather than just looking at 48  
3 hour mortality of 72 or whatever, just having kind of binary  
4 data like that, by looking at these social groups, measuring the  
5 number of individual focal bees that have been treated or not  
6 treated, and as they die over time, that facilitates the ability  
7 to look at survivorship and analysis with survival statistics.

8 So in essence, that's adding that temporal  
9 component to these types of bioassays. So rather than just  
10 taking a cross section in time, you're looking over the course  
11 of time and being able to make important inferences based on  
12 that. So, if you were to just look at 48 hours or really any  
13 one time point, that would be a unit dimensional means of  
14 assessing that. But there are multiple ways that one can arrive  
15 at those same kinds of mortality percentages at that time point.

16 So looking at the curves and the shapes of the curves can be  
17 incredibly informative. That's what is captured by survival  
18 statistics.

19 So, that's one outcome that I think would be  
20 facilitated with really not much additional work in these  
21 screening bioassays to measure the entire lifespans of  
22 individuals and these small in vitro groups, but then  
23 quantifying the slopes of mortality over time would give very,  
24 very informative information, we believe. Again, this has been  
25 done in many studies in this same capacity. So, this is fairly  
26 well documented in the literature.



1                   Because the question also addresses  
2 specifically the effects on queens, and I totally agree that  
3 this is incredibly important. Queens, of course are the sole  
4 reproductive females within colonies. They're kind o the  
5 carrier of all the genetic material in the colony and therefore  
6 a real focal point. Moreover, just on a side note, in the  
7 apiculture industry, there are a lot of reports about problems  
8 with queens, although those problems are quite nebulous. It's  
9 very hard to define them and to pin them down, but there seems  
10 to be consistent issues in the management practices when it  
11 comes to queens. Given their importance to the social  
12 super-organism of honey bee colonies, it seems to be pretty  
13 paramount to try and quantify that phenotype and effects on the  
14 queen phenotype.

15                   There are ways of doing that, of looking at  
16 quantifying queen reproductive potential, not just queen  
17 fecundity that is egg-laying rate, but other aspects of queens  
18 ability to keep the colony healthy, productive and well  
19 populated. I won't go over all of those different measures that  
20 can be done, but let's just suffice it to say that there are  
21 more logical measures and then reproductive measures that could  
22 be done looking at things like the stored sperm count of the  
23 queens, the viability of sperm, ovarial number, egg-laying  
24 rates, all of those types of things.

25                   How those many different characters translate  
26 to the ultimate colony phenotype however, it not entirely known



1 and may be difficult to make extrapolations on measuring those  
2 characters in an individual queen to the ultimate colony  
3 phenotype. So, in that sense, at a Tier I level, it may be very  
4 difficult to be able to quantify effects of particular compounds  
5 on queens and reproductives, the drone as well. So, it might  
6 really necessitate Tier II levels in order to be able to truly  
7 elucidate effects on queens and drones. That's something that  
8 we will get to in later questions as well.

9           Some other potential measurements and  
10 endpoints that the discussants brought up on these in vitro  
11 bioassays is quantifying immunocompetence, effects of compounds  
12 on abilities of individuals to withstand disease or other  
13 challenges. While this is being studied quite heavily in the  
14 literature, it was pretty well agreed that we still don't have a  
15 very good cause effect understanding of a lot of these  
16 measurements such as phenoloxidase activity, encapsulation  
17 response, expression of antimicrobial peptides, those types of  
18 things. They can be done, but we don't understand the full  
19 relationship to the end phenotype. So, while it might be  
20 interesting to keep those in mind, it be premature to include  
21 them in a high throughput endpoint at this point.

22           As was mentioned earlier just a few moments  
23 ago, other endpoints that could and probably should be  
24 considered are changes in behavior, which can be again, very  
25 difficult to quantify and the present their own challenges. But  
26 in a social organism such as honey bees, they are paramount.



1 They are everything. So, looking at the super-organism cannot,  
2 almost by definition, ignore effects on behavior.

3 So, measuring simple behavioral bioassays such  
4 at motility, social interactions, triple axis, other behavioral  
5 changes can be and have been, in the literature, documented. We  
6 also feel that in order to assist this type of data collection  
7 in a repeatable high throughput measure that there is a real  
8 need for technologies that could capture and automate that type  
9 of data collection. And, we know of several initiatives that  
10 are trying to take video recordings of social insect colonies  
11 and trying to automate the complexities of a lot of the social  
12 insect interactions that are going on. So, there ought to be a  
13 real premium on developing those types of technologies in order  
14 to incorporate them into here, but again, at this point in time,  
15 it might be premature.

16 One behavioral bioassay that is well  
17 articulated in the study for decades now is and also discussed  
18 in the White Paper, so I won't go too much into it, but that's  
19 the PER and learning conditioning bioassays. This is proboscis  
20 extension reflex, which incorporates the natural instinct of  
21 honey bee worker to stick out her tongue in response to her  
22 antenna being touched by a small droplet of sucrose solution.  
23 That reflex can then be conditioned and paired with the stimulus  
24 such as an odor or tactile response so that over a few trials of  
25 that association tested individuals will then immediately stick  
26 out their tongues if they are exposed to that same odor. That



1 is a means by which researchers can quantify learning in their  
2 ability to make this association.

3 There is a very rich literature on using PER  
4 for learning and conditioning. These are also potential  
5 endpoints that can be incorporated and harnessed as part of a  
6 Tier I assessment.

7 **DR. DANIEL SCHLENK:** Okay. Dr. Berenbaum?

8 **DR. MAY BERENBAUM:** Just two additional  
9 comments. One is drones represent a significant data gap.  
10 They're sort of literally the forgotten men here. Even though,  
11 they may seem nonessential most of the time, they do serve as a  
12 vital and irreplaceable function in colony life. There's not  
13 much known about their toxicology.

14 The other comment is that just to point out  
15 that the proboscis extension reflex represents a particular type  
16 of learning, repetitive learning, and that's only a component of  
17 the incredibly rich behavioral repertoire of honey bees. So,  
18 it's a proxy maybe. So we have a lot of links to make. First  
19 per has to be linked to in general to colony phenotype and in  
20 addition, its place within the role of very complex behavioral  
21 repertoire of honey bees and needs to be considered.

22 **DR. DANIEL SCHLENK:** Thank you. Dr. Ostiguy?

23  
24 **DR. NANCY OSTIGUY:** I concur with Dr. Tarpy  
25 and Dr. Berenbaum.

26 **DR. DANIEL SCHLENK:** Okay. Dr. Fefferman?



1                   **DR. NINA FEFFERMAN:** Hi, sorry, I feel like  
2 I'm talking a lot. The one thing about queens that I don't see  
3 discussed and it could be completely irrelevant and this is a  
4 toxicology question - is either her pheromone production  
5 directly or the interpretation of pheromones by workers with  
6 higher toxicity at a sublethal effect? There's been a lot of  
7 debate in the literature about who is controlling what happens  
8 because of queen pheromone, but it doesn't really matter if that  
9 gets disrupted. Then that's a huge disruption in colony health  
10 and colony reproduction. So, I have no idea if there're  
11 appropriate assays for this, but my learned colleagues certainly  
12 know if this is an appropriate question to add and if so, how to  
13 measure it if it can be measured.

14                   **DR. DANIEL SCHLENK:** Yes, Dr. Pettis?

15                   **DR. JEFF PETTIS:** There is some recent  
16 literature - recent papers in the literature about the effect of  
17 disease on pheromone production, and probably you might expect  
18 the same thing with some pesticide exposure. Bioassays might  
19 include things like new behavior, the number of bees attending  
20 the queen, things like that, so there are bioassays that could  
21 be looked at.

22                   If I may, I'd just like to comment a bit on  
23 what Dr. Tarpy said about how to look at queen - or how to  
24 incorporate queens into Tier I. I think it's difficult because  
25 of the reports that we are getting of queen failures. I think  
26 it behooves us to try to look at ways to incorporate it. As I



1 pointed out earlier, the queen, we believe and we don't have  
2 good numbers on it, but in effect, they process much more food  
3 than any other worker or drone in the colony because of their  
4 high egg laying rate. So, there could be some biomagnification  
5 of whatever residue is present in the colony.

6 I can think of a couple of ways. With your  
7 group dynamics, it would be possible to just simply introduce  
8 queens into that environment. I don't know if it's practical to  
9 watch behavior, but certainly you could introduce queens and  
10 then see compared to control queens if there was any direct  
11 effect in a Tier I test. So, just adding queens the workers  
12 that are in those Tier Is, it's a possibility.

13 Dave pointed out some of the things we look at  
14 in queen health, such as sperm viability. The queen stores the  
15 sperm, so we can actually measure after a certain period of time  
16 of exposure of the viability of sperm within the queen. I  
17 believe there are issues with queens become drone layers. So,  
18 they either run out of sperm or the sperm is dying. That seems  
19 to be a common theme of late. So, we have bioassays for that as  
20 far as ways of looking at queens.

21 Lastly without sacrificing the queen, you can  
22 actually take a fecal sample from the queen and that might be  
23 telling. You take and analyze the fecal material. But again,  
24 usually at the end of a bioassay, you just sacrifice the queen  
25 and look at a number of parameters. I will say that while we're  
26 doing a lot of this, the methods and the standardization, I



1 don't this is there at this point.

2 **DR. DANIEL SCHLENK:** Yes, Dr. Hunt?

3 **DR. GREG HUNT:** Hi. I'm just willing to bring  
4 up another potential behavioral endpoint, which is homing  
5 ability. There have been a number of recent studies looking at  
6 pesticide effects on bees' ability to find their way home. You  
7 can train them to a feeding station and then take them a  
8 distance to the side to make it a little bit more challenging to  
9 see who gets home. And, this has some - it's very important for  
10 the bees to be able to find their way home, obviously.  
11 Otherwise, they're toast. And it has some advantages over the  
12 PER assay because with the learning assays, it depends on the  
13 bees' sort of motivation, their inherent response, threshold to  
14 sucrose and things of that sort.

15 **DR. DANIEL SCHLENK:** Yes, Dr. Tarpy?

16 **DR. DAVID TARPY:** This is Dave Tarpy again.  
17 So, Greg, I totally agree with that of course. It makes a lot  
18 of sense at Tier II and Tier III, but these being questions  
19 about Tier I, would you think that there would be a good proxy  
20 at the Tier I level that would be able to kind of quantify  
21 homing behavior, kind of, you know, in an in vitro setting?

22 **DR. GREG HUNT:** No, I guess I was getting  
23 ahead of the game there, but I'm glad you pointed that out.

24 **DR. DAVID TARPY:** Well, then bring it up again  
25 with the Tier II. I think that will be - because that is  
26 obviously important.



1                   **DR. DANIEL SCHLENK:** Yes, Dr. McManaman?

2                   **DR. JAMES MCMANAMAN:** So, this is going to  
3 come up in the next session, but I just want to ask the panel.  
4 There's a lot of evidence that there is pretty definitive  
5 genetic changes that occur in queens when they become queens.  
6 They occur in the brain. So, I'm just wondering if genetic  
7 markers could be used as a way as an alternative method of  
8 evaluating queen health and the effects of pesticide on her  
9 reproductive ability.

10                  **DR. DAVID TARPY:** This is Dave Tarpy again.  
11 Actually, there's no genetic - genome of workers and queens are  
12 exactly the same. It's just what genes are getting turned on  
13 and off. That's actually a function. There have been genomic  
14 studies that have looked at caste determination and everything  
15 like that, but unless other panel members would like to correct  
16 me on this, I don't know if any of them would be good genetic  
17 markers that would be reliable to really quantify in a  
18 continuous way of some sort of measure for queen quality and  
19 queen reproductive potential, but I could be wrong on that.

20                  **DR. DANIEL SCHLENK:** Dr. James?

21                  **DR. ROSALIND JAMES:** Two comments. One in  
22 response to Jim. I agree with Dr. Tarpy. We're just not there  
23 yet, I think, in understanding. We're starting to do work with  
24 transcript looking at gene expression, but expression levels of  
25 transcriptome isn't necessarily related to what is actually  
26 happening physiologically and it's complex. We're just



1 beginning to look at that. I don't think it's anywhere near  
2 being ready as some sort of a screening tool.

3 And I had another comment. I was waiting for  
4 some of the queen discussion to die down because I want to  
5 change the subject. You can go back to queens if you want now  
6 that I have the mic. You asked for other endpoint measurements  
7 and we'll probably talk about this a little bit more later when  
8 we get in - you sort of re-asked the same questions in different  
9 ways. So we'll get to this more later, but in part A, we talked  
10 a little bit about using longevity, using these bioassays that  
11 are run for a longer time and one advantage to doing that. And  
12 I'm speaking now about larval bioassays instead of adults as you  
13 can look at development time.

14 So, I think later we'll talk a little bit more  
15 about alternative bioassays, but one of the things you can do  
16 are these larval bioassays and then run them out to some  
17 biological endpoint such as cell capping or pupation, and then  
18 measure how long it takes to get to that stage for each  
19 individual. This would give you another endpoint measurement in  
20 addition to mortality.

21 **DR. DANIEL SCHLENK:** Okay. Any other  
22 comments? I didn't hear growth mentioned at all. Is that an  
23 endpoint? Because that's mentioned in the question. Mr.  
24 Pistorius?

25 **MR. JENS PISTORIUS:** One comment that is also  
26 related to charge question 9, I apologize if I may be a bit



1 early in this. I think growth could be included in the future  
2 coming Aupinel test method or OECD guidelines, which is  
3 developed and regarded in the Tier I, although it needs to be in  
4 short, additional handling of the larvae does not affect  
5 mortality, which is a bit critical in this test. I think this  
6 is the only Tier I test method which has been or is under  
7 validation that would actually cover growth on a Tier I.

8 **DR. DANIEL SCHLENK:** Dr. James?

9 **DR. ROSALIND JAMES:** Hopefully this isn't just  
10 reiterating the same thing, but the larvae are extremely  
11 delicate and you can't really pick them out and weigh them  
12 regularly. At the end, perhaps you could weight them, but  
13 growth is difficult to measure for the larvae. And then the  
14 adults, you are not going to have growth occurring. So, this is  
15 one reason way we use development time rather than growth.

16 **DR. DANIEL SCHLENK:** Yes, Dr. Fefferman?

17 **DR. NINA FEFFERMAN:** Although it is - well,  
18 I'm sure everyone meant individual growth, the rate of change of  
19 the size of the colony after mating or once the queen starts egg  
20 laying, not just a measurement of fecundity but a successful  
21 rate of change - that ramp up, the differential there might  
22 actually be a meaningful colony health measure.

23 **DR. DANIEL SCHLENK:** Yes, Dr. Ostiguy?

24 **DR. NANCY OSTIGUY:** One difficulty with that  
25 is there are tremendous strain differences. So, if you're going  
26 to ask whether or not or how fast a colony builds, you'd better



1 standardize for which type of honey bee you're using.

2 **DR. DANIEL SCHLENK:** Mr. Pistorius?

3 **MR. JENS PISTORIUS:** Especially regarding  
4 growth, we would like to discuss this also in the Tier II and  
5 Tier III tests because they are very reliable and very  
6 well-suited methods available for measuring growth.

7 **DR. DANIEL SCHLENK:** Okay. Any other comments  
8 on this section? Dr. Delclos?

9 **DR. KENNETH DELCLOS:** Question - now I think  
10 that in the first part A, I think we are talking about using a  
11 single dose. How critical is the timing of the dose for these  
12 various - I mean, I don't know anything about the development of  
13 metabolic capacity. I don't hear much about that toxicokinetics  
14 discussed in here, but also in the response if you're looking at  
15 a specific response, how tight a window - is there an apriority  
16 way of choosing one that should be administered?

17 **DR. DAVID TARPY:** This is David Tarpy. I  
18 don't think there is an apriority way, but often times in these  
19 bioassays, they're done by newly closed workers, newly emerged  
20 workers as they're placed into these experimental arenas, but  
21 it's true. There may be differences in the time of application  
22 on their effects.

23 **DR. DANIEL SCHLENK:** Okay. Any other comments  
24 for question 8? Okay. I'm going to go to the Agency -  
25 questions of clarification? Mr. DeCant?

26 **MR. JOSEPH DECANT:** Yeah, this is Joe DeCant



1 from EFED. I just have a question. It sounds like - thank you  
2 to the Panel. We got some good feedback in terms of the number  
3 of different endpoints. I think some of the endpoints like Dr.  
4 Tarpy, you mentioned about immunocompetence. It sounds like  
5 some of the endpoints are quite there and aren't quite there and  
6 aren't quite developed yet to be able to include into some of  
7 our studies. I also want to ask a clarification question - some  
8 of the endpoints that were mentioned like the PER Assay - I know  
9 that we are trying to link these endpoints with assessment  
10 endpoints. So survival, mortality and to what extent do you  
11 think some of these measurement endpoints are actually linked to  
12 our assessment endpoints?

13 **DR. DANIEL SCHLENK:** Dr. Tarpy?

14 **DR. DAVID TARPY:** This is Dave Tarpy again. I  
15 would argue all of them to greater or lesser degrees. Again,  
16 adding that extra layer of biological complexity, that is the  
17 colony, inherently brings in all of these different aspects. I  
18 think logistically in measuring them in a repeatable,  
19 quantifiable way is really a difficulty. And as you're correct  
20 at that immunocompetence measures may not be as well elucidated  
21 to be able to make those connections to the social level yet,  
22 but it is definitely something to keep in mind. Some of the  
23 other bioassays like the PER are more robust and standardized.

24 So, we bring all of them up as a means of  
25 keeping them in mind as to complexity of dealing with the social  
26 organism rather than a solitary one, but that there is an



1 understanding that just based on feasibility that not all of  
2 them are going to be incorporated or at least right away.

3 **DR. DANIEL SCHLENK:** Dr. Berenbaum?

4 **DR. MAY BERENBAUM:** With that said, I think  
5 it's important to - at least one component of your question is  
6 the linkage to colony phenotype. And, I don't even think for  
7 proboscis extension reflex that there is a bulletproof linkage.  
8 So, there is another data gap.

9 **DR. DANIEL SCHLENK:** Yes, Mr. Pistorius?

10 **MR. JENS PISTORIUS:** I still think it is very  
11 difficult to actually link the findings of the PER Test to the  
12 realistic situation that bee colonies are encountered with. I  
13 would like to point out that there is an area of major need for  
14 research to make this link to actually establish a link and only  
15 very limited number of studies that have looked into it, but  
16 with those for instance, we could find some effect in PER, we  
17 could not find any relation as to foraging behavior or to honey  
18 flow or honey yield of the colonies. But again, there is so  
19 much limited data available on how to link those two that I  
20 think we're a little bit unsure about this.

21 **DR. DANIEL SCHLENK:** Dr. Tarpy?

22 **DR. DAVID TARPY:** This is Dave Tarpy again. I  
23 failed to mention last time, and I agree with all of the points  
24 that were raised there is because of these difficulties and  
25 linkages to full colony phenotypes, it places a real premium and  
26 a priority on Tier II and III bioassays, and that it is almost



1 inherent to study a super-organism at the level of that  
2 super-organism, which is the colony. So, it's incredibly  
3 difficult to use Tier I in vitro tests to truly adequately  
4 capture the entire biology at the colony level. So that's an  
5 inherent difficulty seen here, which emphasized Tier II and Tier  
6 III bioassays for these screenings.

7 **DR. DANIEL SCHLENK:** Okay. Dr. Ostiguy?

8 **DR. NANCY OSTIGUY:** I'm going to disagree a  
9 little bit with some of the comments. I actually think the PER  
10 does provide some very useful information that can be related  
11 then to the full colony in the same way that we evaluate muscle  
12 rechemical exposure, et cetera, on human learning ability that  
13 we then apply to a population. So, if you adversely affect  
14 individuals within the group, which is what we're looking at  
15 with honey bees, you are going to at some point impact the whole  
16 group. So to me, there's the connection, you know. The  
17 individuals do make up the whole. And if we interfere with the  
18 ability of individual bees to learn, then we interfere with the  
19 whole. So, what we might need to do with the PER is set a  
20 particular standard for the degree of learning inhibition rather  
21 than saying we can't use the PER as a Tier I screen to try to  
22 get at some of that some of that social insect interaction.

23 **DR. DANIEL SCHLENK:** Okay. Dr. Fefferman?

24 **DR. NINA FEFFERMAN:** We'll talk more about  
25 this at the end of everything, but I think that kind of question  
26 is exactly the sort of thing where modeling can help.



1                   **DR. DANIEL SCHLENK:** Dr. Pettis?

2                   **DR. JEFF PETTIS:** Just in regard to the PER  
3 and other tests that we might run, I agree that the PER has been  
4 vetted well as far as demonstrating effects, but I think I would  
5 have more confidence in something like Dr. Hunt suggested in  
6 these returning forager assays, but it may not be exactly a Tier  
7 I. It may be closer to a Tier II test where you actually dose  
8 foragers at realistic exposure levels and then ask them to  
9 return to the hive. I think there have been several papers on  
10 that of late. They seem very convincing that realistic field  
11 exposure can disrupt returning forager behavior. You can see  
12 that that can have direct effects on the colony.

13                   **DR. DANIEL SCHLENK:** Okay. Dr. Klaine? Dr.  
14 Ostiguy?

15                   **DR. NANCY OSTIGUY:** I actually agree, but I  
16 think Tier I -- using PER as a Tier I would then trigger us to  
17 do that field returning ability. So, the PER could be set  
18 sufficiently conservative so that if we need to, we can then  
19 check to see if it really has that effect on the whole group.

20                   **DR. STEPHEN KLAIN:** This is Steve Klaine with  
21 Clemson. Thinking about this on a larger scale in terms of  
22 trying to move from a single organism to a more complex  
23 super-organism, it seems to me that any one behavior may not get  
24 us all the way, but that a bank of these behaviors may give us a  
25 lot of insight into the whole hive health by using single  
26 organisms. So, I think it ultimately - and I agree with the



1 discussions last night - we're not there yet and what you said  
2 today, but I think that's where we need to be if we're going to  
3 start making predictions on hive health.

4 **DR. DANIEL SCHLENK:** Anyone else to weigh in  
5 on this one? No? I hear some discussions back there. Do you  
6 guys need some more clarification or are we.

7 **MR. JOSEPH DECANT:** This is Joe DeCant from  
8 EFED. Could you elaborate a little bit on the bank of behaviors  
9 that you mentioned about and what type of behaviors will be  
10 helpful? I know Dr. Pistorius, you mentioned about some of the  
11 behavioral effects that are observed in the OECD Guidelines, and  
12 so if you could clarify or elucidate on those behaviors.

13 **MR. JENS PISTORIUS:** Well there are a number  
14 of described categories of behavioral abnormalities, and it is  
15 quite difficult to now put really into a frame when this would  
16 actually trigger a real additional concern. But for instance,  
17 during my work, I was encountered with one substance that had  
18 doses clearly below the lethal doses at a number of dosings,  
19 also in very low dosings, in different laboratories, repeatable  
20 effects, severe effects on behavior of very agitated, very  
21 nervous, trembling, shaking, all this. So, I would just like to  
22 point out is that it is important that the assessed behavioral  
23 abnormalities are reported by the contractors and that the risk  
24 assessor also regardless of any trigger values still has the  
25 flexibility to say, on the basis of these concerns, we have to  
26 go to a higher tier testing and do additional testing. Does



1 that answer your question?

2 **DR. DANIEL SCHLENK:** Any other questions, Mr.  
3 DeCant? Oh, I'm sorry, Dr. Klaine has something to add here.

4 **DR. STEPHEN KLAINE:** Yeah, I was just - you  
5 talked about the bank - I mentioned a bank of behaviors. I  
6 think there are a lot of them that maybe aren't standardized but  
7 at least in my discussions with my bee colleagues, have really  
8 been intriguing. The discussion we had last night on the  
9 nursing, the in vitro nursing assay, where you could - that's  
10 another behavior that you can quantify, because in my mind, at  
11 least, it's not one behavior that describes the health of the  
12 hive, but its culmination of all these behaviors that make these  
13 hives work. So, if you can capture, quantify three or four of  
14 them, you might get a lot better insight into bee health or hive  
15 health.

16 **DR. DANIEL SCHLENK:** Okay. Anyone else? Let  
17 me go back to Mr. DeCant. Do you have any other questions of  
18 clarification.

19 **DR. JOSEPH DECANT:** No, I think we're good.

20 **DR. DANIEL SCHLENK:** Great. Okay. Let's go  
21 ahead and move onto question 9. Want to read letter A into the  
22 record for us?

23 **MR. JOSEPH DECANT:** Joe DeCant, EFED.  
24 Question 9, subpart (a) - please comment on the extent to which  
25 the Aupinel et al. in vitro method serves as an appropriately  
26 conservative estimate of Tier I acute oral exposure of honey bee



1 larvae to pesticides, given differences in this test design from  
2 actual in-hive exposure conditions, for example during the first  
3 three days of the larval development stage, larvae consume royal  
4 jelly and brood food, and the uncertainty regarding the extent  
5 to which larvae rely exclusively on pollen or nectar as opposed  
6 to royal jelly or brood food.

7 **DR. DANIEL SCHLENK:** Our lead discussant for  
8 this question is Dr. Berenbaum.

9 **DR. MAY BERENBAUM:** Determining toxicity of  
10 pesticides to larvae using in vitro testing is complicated by  
11 several factors including possible genetic differences among  
12 colonies and difficulties synchronizing larval age, both of  
13 which were mentioned by Aupinel and also included in the White  
14 Paper. In addition as yet uncharacterized attributes of royal  
15 jelly, brood food, honey and bee bread may be relevant to  
16 pesticide tolerance in larvae.

17 Royal jelly, for example, contains substances  
18 that inhibit DNA methylation and histone deacetylase activity,  
19 also promote gene expression and similarly honey and bee bread  
20 constituents contain, including phytochemicals that can regulate  
21 detoxification pathways in worker bees at least. These factors  
22 have the potential to alter responses to pesticides, but without  
23 more complete understanding of how the composition of food  
24 affects toxicity or bioavailability despite compound, it's  
25 difficult to say conclusively that the method is conservative,  
26 although available information points in that direction.



1                   One specific concern is that the proposed  
2 method for testing larvae is based on pesticide levels in pollen  
3 and nectar administered to day-5 larvae. Day-5 larvae destined  
4 to be queens eat royal jelly. If royal jelly has 1/100th the  
5 level of pesticides contaminants the nectar and pollen, test  
6 conducted in Tier I using maximum concentrations of pesticides  
7 based on worker exposure will be much higher than those that are  
8 experienced by queen larvae.

9                   So, in terms of toxicity, this approach is  
10 reasonable, but if caste determination and hive viability are  
11 concerns, then these pesticide amounts might not be appropriate.

12       As noted earlier too, the assumption that royal jelly  
13 concentrations of pesticides are 1/100th those in other bee  
14 foods, those estimates are based on one published and one  
15 unpublished study. So, that underlines that assumption.

16                   So, the impact of larval diet on pesticide  
17 detoxification represents a significant data gap. I forgot to  
18 introduce my comments by saying I am extremely grateful that the  
19 EPA is undertaking this effort and also to point out that the  
20 first indication that pesticides used for agricultural purposes  
21 have nontarget impacts on bees goes back to 1895, and the fact  
22 that we are now a century later talking about major data gaps, I  
23 think is unsettling at the least, and that's one reason I'm very  
24 grateful to the EPA for convening this Panel.

25                   Okay. So, data gap - the artificial diet used  
26 for bioassay contains glucose and fructose as proxies for honey,



1 but honey is substantially more than just carbohydrates.  
2 Phytochemicals of honey may well influence pesticide toxicity  
3 and metabolism as they do in other herbivorous insects. There  
4 is a rich literature in the effect of phytochemicals and  
5 detoxification in other herbivores. But in larval bees, it's  
6 essentially nonexistent.

7 More generally, the concept of an acute dose  
8 for a larva may be biologically difficult to interpret. Unlike  
9 workers who may experience one-time encounters through pesticide  
10 drift outside the hive, larvae mature continuously provide food  
11 that has been already processed by workers seem less likely to  
12 have acute exposures more over because larvae are confined  
13 during their development, they're subject to continuous contact  
14 with wax, which if contaminated is likely to be a source of  
15 toxicity that is not directly assessed by the proposed methods.  
16

17 Contact bioassays would be very useful in this  
18 context, particularly using pupae just for operational and  
19 logistic reasons and also there are precedents for pupal  
20 bioassays of microbial toxicity and the like. I guess those are  
21 my comments.

22 **DR. DANIEL SCHLENK:** Okay. Thanks. Dr.  
23 McManaman?

24 **DR. JAMES MCMANAMAN:** My comments were  
25 incorporated into Dr. Berenbaum's.

26 **DR. DANIEL SCHLENK:** Dr. Ostiguy as well?



1                   **DR. NANCY OSTIGUY:** I concur, my comments were  
2 included by Dr. Berenbaum.

3                   **DR. DANIEL SCHLENK:** Okay. Open to other  
4 Panel members to weigh in on this one. Yep, Dr. James?

5                   **DR. ROSALIND JAMES:** I would have to weigh in  
6 on the larval bioassays. Because I do so many of them myself,  
7 although not with honey bees, but in line with what Dr.  
8 Berenbaum said about the one time acute exposure, one of the  
9 things that we talk about is contaminating all of the food that  
10 you feed to the larvae. So, during the full five-day feeding  
11 period, the bees would be continually fed with pesticide  
12 contaminated food stuff instead of a one-time dose. This would  
13 also get at the question about which age is most sensitive in  
14 the developing larvae, which size increase is quite amazing, and  
15 we don't really know about sensitivity of the different ages.  
16 But I think it would be more realistic to have - they would  
17 continually be fed pesticides.

18                   **DR. DANIEL SCHLENK:** Any other comments? Just  
19 curious on my own side here is would you propose then a  
20 standardized diet that would have the same reference material in  
21 terms of phytochemical input that could be a baseline  
22 standardized diet that would used? Is that something that you  
23 would propose?

24                   **DR. MAY BERENBAUM:** This is May Berenbaum. It  
25 would be ideal. It would be preferable. What constituents  
26 would go into that diet would be probably subject to



1 considerable discussion.

2 **DR. DANIEL SCHLENK:** Okay. Anything else on  
3 A? Oh, sorry.

4 **DR. JAMES MCMANAMAN:** I have a question for  
5 the Panel. This is just out of ignorance - is royal jelly royal  
6 jelly? I mean, are the constituents of royal jelly independent  
7 of strain of kind of bee?

8 **DR. ROSALIND JAMES:** This is Rosalind James.  
9 I'm going to let one of the honey bee folks answer that  
10 question, but I have to add that you do want to make sure that  
11 when you buy the royal jelly it's not all ready contaminated  
12 with pesticides.

13 **DR. DANIEL SCHLENK:** Dr. Pettis?

14 **DR. JEFF PETTIS:** Jeff Pettis. I was going to  
15 say a solitary person commenting on royal jelly, come on Rose.  
16 That was going to be my point was that royal jelly is royal  
17 jelly by and large, but you really have to consider the source  
18 and we've had major issues in doing lab studies and even trying  
19 to do any kind of larval rearing in that we get contamination in  
20 the royal jelly, so that the batch of royal jelly has to be  
21 tested and be standardized and known to be clean before it's  
22 used. Otherwise, you get confounding results.

23 **DR. DANIEL SCHLENK:** Dr. Fefferman and Dr.  
24 Hunt?

25 **DR. GREG HUNT:** I was just about to say that  
26 that was an excellent question. I don't really know what the



1 variability is in royal jelly, but ideally, we would have an  
2 artificial diet. It seems that just doesn't work very well.

3 **DR. DANIEL SCHLENK:** That was Dr. Hunt. Dr.  
4 McManaman? Hold on, let me get Dr. McManaman back.

5 **DR. JAMES MCMANAMAN:** Yeah, so my question was  
6 - you bring up some important points about contamination, but my  
7 question was related to the charge question is that if we're  
8 trying to understand the effects of pesticides and the food can  
9 interfere or affect their responses and royal jelly is a  
10 component of that, especially for queens in early development,  
11 then I think it's important to know whether it really is the  
12 same thing because there are factors in royal jelly that can  
13 influence strain differences.

14 **DR. DANIEL SCHLENK:** Okay. Dr. Fefferman?

15 **DR. NINA FEFFERMAN:** Sorry, thanks. I still  
16 have the same question I had before, which is when you say  
17 contamination in the royal jelly that you're buying, is it  
18 pesticide contamination? Because from listening to the  
19 conversation just a bit ago, I was under the impression that  
20 royal jelly pesticide contamination was really sort of the  
21 lowest level of concern in terms of what they were eating, and  
22 that sounds scary.

23 **DR. DANIEL SCHLENK:** Dr. Pettis?

24 **DR. JEFF PETTIS:** Probably one of the major  
25 contaminants is antibiotics. You can get antibiotic  
26 contamination. Beekeepers use antibiotics in the colony to



1 control bacterial diseases. I think 80 percent or 90 percent of  
2 the world's production of royal jelly comes from China, and so  
3 you can have issues there. You need to really get your royal  
4 jelly from colonies that are managed almost totally organic.  
5 You also can get some miticide compounds that are used by  
6 beekeepers to control mites that get incorporated in the royal  
7 jelly.

8 **DR. DANIEL SCHLENK:** Dr. Ostiguy and then Dr.  
9 Hunt?

10 **DR. NANCY OSTIGUY:** We also need to be  
11 concerned about the possible pathogens that are in that royal  
12 jelly.

13 **DR. DANIEL SCHLENK:** Dr. Hunt?

14 **DR. GREG HUNT:** I was just going to comment  
15 that it might be best to collect your royal jelly fresh and then  
16 just make sure that the controls and the treated get the same  
17 stuff.

18 **DR. DANIEL SCHLENK:** Okay. Any other comments  
19 on 9A? Mr. DeCant, do you have all you need for 9A there?

20 **MR. JOSEPH DECANT:** I do.

21 **DR. DANIEL SCHLENK:** Okay. All right, let's  
22 move on to the second question of 9.

23 **MR. JOSEPH DECANT:** Joe DeCant, EFED, question  
24 9, subpart (b) please comment on the extent to which pesticides  
25 would be more or less bioavailable using the synthetic matrix  
26 relied on for feeding developing bees in this in vitro method.



1 **DR. DANIEL SCHLENK:** Dr. Berenbaum?

2 **DR. MAY BERENBAUM:** Bioavailability will be  
3 influenced by the nature of the compound, larval metabolism,  
4 possibly food composition. Without accurate information about  
5 how these factors influence bioavailability within honey bee  
6 larvae, it's not really possible to really answer this question.  
7 Aupinel et al. 2007 used dimethoate, which is an  
8 organophosphate the highly water soluble. It's not clear how  
9 pesticides with different structures and properties will perform  
10 in this particular synthetic matrix.

11 **DR. DANIEL SCHLENK:** Okay. Dr. McManaman?

12 **DR. JAMES MCMANAMAN:** I have nothing to add.

13 **DR. DANIEL SCHLENK:** And Dr. Ostiguy?

14 **DR. NANCY OSTIGUY:** I concur with what Dr.  
15 Berenbaum has said all ready.

16 **DR. DANIEL SCHLENK:** Anyone else on this  
17 section? Okay. Seems pretty clear to the point. Any  
18 clarification question there Mr. DeCant?

19 **MR. JOSEPH DECANT:** I think we're good.

20 **DR. DANIEL SCHLENK:** I'm hoping so. Okay.  
21 We're going to move on to C please.

22 **MR. JOSEPH DECANT:** Joe DeCant, EFED, question  
23 9, subpart (c) - please comment on the extent to which the  
24 absence of trophallaxis that is the transfer of food or fluids  
25 between colony members, may render larvae more or less  
26 vulnerable to pesticides.



1 DR. DANIEL SCHLENK: And Dr. Berenbaum?

2 DR. MAY BERENBAUM: You thought B was straight  
3 forward. In response to the question, contribution of  
4 trophallaxis to pesticide resistance or susceptibility is to our  
5 knowledge completely and utterly unknown. This represents a  
6 data chasm.

7 DR. DANIEL SCHLENK: Okay. Dr. McManaman?

8 DR. JAMES MCMANAMAN: A very deep chasm, I  
9 totally concur.

10 DR. DANIEL SCHLENK: And Dr. Ostiguy?

11 DR. NANCY OSTIGUY: I have to commend the EPA  
12 for even asking the question, because we truly have no idea.

13 DR. DANIEL SCHLENK: Yes, Dr. Berenbaum?

14 DR. MAY BERENBAUM: Let me add that it's  
15 important. So, it is important to find out.

16 DR. DANIEL SCHLENK: Yes, Dr. Tarpy?

17 DR. DAVID TARPY: And not to beat the dead  
18 horse, but I don't want to move on too quickly. And that is I  
19 think this again underscores the complexities of all of the  
20 social interactions when we're dealing with a super-organism as  
21 opposed to just individual larvae or individual adults.  
22 Everything is interconnected in these types of social systems.  
23 So, these are important questions to ask, but it just  
24 underscores our data gap and the complexity of the social  
25 organism.

26 DR. DANIEL SCHLENK: Dr. James, did you have



1 something?

2 **DR. ROSALIND JAMES:** Well, the only thing I  
3 would add is, of course, increases of spread of pesticides that  
4 is collected and then it may end up getting fed to many more  
5 individuals than originally come into contact with it. In that  
6 way, it would perhaps dilute the pesticide, but spread it out  
7 among more individuals.

8 **DR. DANIEL SCHLENK:** Okay. Any other comments  
9 on C? Mr. DeCant, did you get what you needed on that one?

10 **MR. JOSEPH DECANT:** Yeah, I think we're good.  
11 Thanks.

12 **DR. DANIEL SCHLENK:** All right. Then read  
13 letter D into the record please.

14 **MR. JOSEPH DECANT:** Joe DeCant, question 9,  
15 subpart (d) - please comment on alternative methods for testing  
16 individual larvae that may be appropriate for quantitative use  
17 in a Tier I screening-level assessment.

18 **DR. DANIEL SCHLENK:** Okay. Dr. Berenbaum?

19 **DR. MAY BERENBAUM:** Literature reports of  
20 alternative methods for testing larvae quantitatively are  
21 vanishingly rare. Atkins and Kellum in 1986 treated larvae in a  
22 colony by micropipetting pesticides directly into the cells, but  
23 that might quantify more as a Tier II rather than Tier I. Pupal  
24 bioassays have not been widely used for pesticides toxicity  
25 testing, but they have been used in other contexts, and it seems  
26 that they may be productively employed in this context,



1 particularly given the confounding problems of social context.  
2 I mean, well it said people live alone and die alone - the same  
3 is true for bees, but they also pupate alone. So, there is one  
4 aspect of their life that is independent, more or less  
5 independent of a social context. So, that might be particularly  
6 suitable for Tier I.

7 Let's see - following up on Dr. Tarpy's  
8 suggestions about other endpoints, larval transcriptomes of  
9 honey bees have been described. The existence of such data  
10 provides quantitative measures of effects of toxins on larval  
11 development that might be useful as alternative indicators.  
12 Genetic resources could allow the use of gene expression,  
13 detoxification, antioxidant enzyme activities, senescence traits  
14 as biomarkers, but in fact, the Johnson et al. 2009 suggested  
15 the use of ribosomal RNA fragments as a biomarker, but such  
16 methods would require vetting and validation before they could  
17 be used in Tier I screening.

18 **DR. DANIEL SCHLENK:** Okay. Dr. McManaman?

19  
20 **DR. JAMES MCMANAMAN:** My comments were  
21 incorporated in Dr. Berenbaum's response.

22 **DR. DANIEL SCHLENK:** And Dr. Ostiguy?

23 **DR. NANCY OSTIGUY:** I concur with what's been  
24 said.

25 **DR. DANIEL SCHLENK:** Okay. Other Panel  
26 members? Dr. James?



1                   **DR. ROSALIND JAMES:** I'm going to call on Dr.  
2 Tarpy because last night he mentioned a cage study where you put  
3 the brood with a few workers. We're getting to that? All  
4 right.

5                   **Dr. DANIEL SCHLENK:** I think that's for  
6 question 10 right? I think they're going to get to that later.  
7 Anyone else for D? New alternatives? Okay. Mr. DeCant, any  
8 questions of clarification?

9                   **MR. JOSEPH DECANT:** Joe DeCant, EFED. Maybe  
10 one question, Dr. Berenbaum, you mentioned about pupal  
11 bioassays. Do you have references for these protocols? Have  
12 they been around for a while?

13                   **DR. MAY BERENBAUM:** I also defer to Dr. Tarpy.

14                   **DR. DAVID TARPY:** This is Dave Tarpy. I don't  
15 have them off the top of my head. All I know is that there are  
16 - well, we're writing a paper right now looking at that. I know  
17 that many of those bioassays are also being utilized in the  
18 Beltsville Lab. So, we can dig those up for you for sure, yeah.

19  
20                   **DR. DANIEL SCHLENK:** Yeah, it sounds like if  
21 you could provide references for some of those, it would be  
22 good. Dr. James?

23                   **DR. ROSALIND JAMES:** But this could only be a  
24 contact toxicity bioassay.

25                   **DR. DANIEL SCHLENK:** Okay. All right, let's  
26 go ahead and move onto letter E.



1                   **MR. JOSEPH DECANT:** Joe DeCant, EFED, question  
2 9, subpart (e) - typically acute toxicity tests are concluded  
3 between 48 and 96 hours. Please comment on the appropriate  
4 duration of toxicity tests for assessing acute toxicity to  
5 individual larval and adult bees.

6                   **DR. DANIEL SCHLENK:** And Dr. Berenbaum.

7                   **DR. MAY BERENBAUM:** The problems interpreting  
8 larval survival over a two to four day period is a metric of  
9 toxicity. The four day endpoint represents 80 percent of the  
10 period of active feeding. So, distinguishing between acute and  
11 chronic becomes difficult. Some agents induce toxicity due to  
12 the larval development, which would be over looked in the  
13 proposed approach. In general, biological transitions such as  
14 pupation provide a more reliable consistent and interpretable  
15 endpoint. Time to pupation, pupation rate and pupal weights are  
16 more biologically meaningful. Queens and drones can be assessed  
17 separately.

18                   Such tests however, have not yet been vetted  
19 thoroughly. Assessing acute toxicity over seven days for the  
20 present time will allow the EPA to harmonize in the national  
21 efforts at the OECD level until a satisfactory standard pupation  
22 assay has been developed, but developing such a test should be a  
23 priority. Among other things, these assays will be more  
24 sensitive to pesticide impacts on growth rate and development,  
25 such as IGRs.

26                   **DR. DANIEL SCHLENK:** Thank you. Dr.



1 McManaman?

2 **DR. JAMES MCMANAMAN:** I concur.

3 **DR. DANIEL SCHLENK:** Dr. Ostiguy?

4 **DR. NANCY OSTIGUY:** I concur.

5 **DR. DANIEL SCHLENK:** Other Panel members?

6 Nothing further to add? Wow, that's a shocker. Okay. Oh  
7 sorry. Kiss of death on there. Mr. Pistorius?

8 **MR. JENS PISTORIUS:** Thank you. One thing to  
9 add regarding the in vitro test, I think it is important that  
10 also additional standards are testing. Now in the reference, it  
11 has mainly mentioned dimethoate. Also fenoxycarb would be  
12 necessary, so that should be principally considered in the  
13 guidelines.

14 **DR. DANIEL SCHLENK:** Okay. Anyone else?  
15 Okay. Mr. DeCant, any questions of clarification?

16 **MR. JOSEPH DECANT:** No more questions.

17 **DR. DANIEL SCHLENK:** Oh, hold on. Ms. Garber  
18 has something.

19 **MS. KRIS GARBER:** Kris Garber. Dr. Berenbaum,  
20 you indicated that it be more relevant to use the seven-day  
21 duration? And that's what we've been calling the chronic  
22 toxicity test? So does that mean that you're suggesting the we  
23 kind of eliminate the ideas of acute and chronic for larvae and  
24 just have one, kind of one risk quotient, one toxicity test?

25 **DR. MAY BERENBAUM:** This is May Berenbaum.  
26 This is the conclusion we reached yesterday. It's just



1 biologically hard to interpret and acute exposure of a larva.  
2 So, given the relative return on investment in terms of, you  
3 know, information that's relevant to deciding about risk and on  
4 whether to proceed to Tier II. The general consensus, at least  
5 among the discussants last night is that maybe just time to  
6 pupation, that interval is more biologically informative. I  
7 don't know if anyone else wants to weigh in here.

8 **DR. DANIEL SCHLENK:** Mr. Pistorius?

9 **MR. JENS PISTORIUS:** To now, the in vitro  
10 method is described especially for the period of egg or young  
11 larvae until the phase of pupation. But by doing this and  
12 ending there, you may get a toxicity for the larvae when they  
13 are white, but we know for some substances like fenoxycarb,  
14 which has been shown to be critical under practical conditions  
15 that those affect after pupation before emergence. So, by just  
16 looking at the first seven days, you will miss this effect and  
17 this has been an effect, which has been critical in practices.  
18 This is not developed yet in the in vitro method. It has not  
19 been validated. So, this is a major issue that should be pushed  
20 further immediately, that this test can be extended. Instead,  
21 there are several endpoints in this test. One would be the  
22 seven-day exposure and the larval mortality. Then the second  
23 one is the time until hatch as the endpoint, the second one.

24 **DR. DANIEL SCHLENK:** Okay. Dr. Ostiguy?

25 **DR. NANCY OSTIGUY:** Basically, at least my  
26 understanding of the conversation was that you would get the



1 information about acute toxicity without actually having to run  
2 a separate test. So, you then would be able to, right now,  
3 what's being proposed and certainly is acceptable because it's  
4 been tested and is reliable, is measurements to the seven days.  
5 Then, as soon as we get information that allows us to take the  
6 larva to pupation and then to emergence, those would be good  
7 endpoints to strive for. So, method development is needed for  
8 the latter two pieces.

9 **DR. DANIEL SCHLENK:** Okay. Dr. James? Yep.

10  
11 **DR. ROSALIND JAMES:** I'll just add, just in  
12 case it's not clear. If you're going to do development time,  
13 you'll be measuring the larvae every single day anyway. So,  
14 you'll still have your 48 and 96 hour mortality measures in  
15 there because you need to do that for your survival time  
16 analysis.

17 **DR. DANIEL SCHLENK:** Any other questions from  
18 the Agency? We're good? Okay. Let's go ahead and take a break  
19 right now. We will convene at 20 till.

20 (Brief recess.)

21 Mr. DeCant, you want to read in question  
22 10(a)?

23 **MR. JOSEPH DECANT:** Joe DeCant, EFED, question  
24 10, subpart (a) - please comment on the conclusion that adequate  
25 procedures have not been sufficiently developed and validated  
26 for assessing chronic toxicity to individual bees in a risk



1 assessment context.

2 **DR. DANIEL SCHLENK:** Dr. Berenbaum?

3 **DR. MAY BERENBAUM:** The concept of the  
4 individual adult bee presents a challenge to assessing toxicity  
5 in *Apis mellifera* because of the importance of the social  
6 context, age-related polyethism over time is circumvented in the  
7 absence of a social medium in foragers are hard-to-keep alive  
8 individually. However, exposing emerging workers for 10 days at  
9 low doses with known ages of bees has been attempted according  
10 to OECD 213 with some success.

11 Cage brood tests and broodless bee cup tests  
12 may also be employed productively in this context. So, it's  
13 important to emphasize that, although there may not be at the  
14 moment entirely reliable and interpretable assays assessing  
15 chronic toxicity is nonetheless remains very important.

16 **DR. DANIEL SCHLENK:** Okay. Thanks. Our next  
17 discussants, Dr. - where am I - Dr. James, yes?

18 **DR. ROSALIND JAMES:** That's me, Rosalind  
19 James. I concur. I would just add that we had some discussion  
20 about this on the previous question about possible chronic tests  
21 that could be done in lieu of doing actually the acute tests.

22 **DR. DANIEL SCHLENK:** Dr. Pettis?

23 **DR. JEFF PETTIS:** I concur with what's been  
24 said.

25 **DR. DANIEL SCHLENK:** Dr. Ostiguy?

26 **DR. NANCY OSTIGUY:** I concur.



1                   **Dr. DANIEL SCHLENK:** Other panel members want  
2 to weigh in on this one? Okay. Mr. DeCant, do you have what  
3 you need on that one? Okay. Let's move onto 10B. Want to read  
4 that in? I have to admit, that was Keith Sappington's  
5 recommendations.

6                   **MR. JOSEPH DECANT:** Joe DeCant, EFED -- please  
7 comment on the potential use of the 10 day adult worker and  
8 seven-day in vitro larval toxicity tests discussed in the White  
9 Paper for assessing chronic toxicity once these methods are  
10 fully vetted.

11                  **DR. DANIEL SCHLENK:** Dr. Berenbaum?

12                  **DR. MAY BERENBAUM:** The optimal design, we  
13 feel, for a larval in vitro test would be one that encompasses  
14 the entire active feeding period through pupation with time to  
15 pupation, pupal weight, percent pupation, as quantifiable and  
16 interpretable measurements of pesticide impact. Such a test can  
17 provide data on both acute and delayed responses. It goes  
18 without saying, but I will nonetheless say, that such testing  
19 requires vetting of course.

20                         With respect to adults, the use of a 10-day  
21 viability test for adult bees, although it does present some  
22 interpretation challenges, nonetheless has a virtue of allowing  
23 for harmonization with ongoing efforts in Europe.

24                  **DR. DANIEL SCHLENK:** Dr. James?

25                  **DR. ROSALIND JAMES:** Yes please do them. I  
26 don't know how else to emphasize that the 48 hour, 96 hour tests



1 are a great frustration to anybody working with bees and  
2 toxicity is how come EPA looks at such a short time period? I  
3 believe perhaps Dr. Pistorius can comment on - there are some  
4 standard OECD tests already available for the seven-day test.

5 **MR. PISTORIUS:** Thank you. If I may answer  
6 directly, there is no real OECD test guideline available for the  
7 10 day test, but what is done and what is asked for in special  
8 cases with the concern of especially insecticides is that the  
9 OECD 213 is used and the protocol is used accordingly to OECD  
10 213, which means basically that all the test conditions, like  
11 how many bees per cage in all this is used accordingly, but the  
12 test is extended to 10 days. The only difference is that  
13 usually freshly emerged bees are taken, so this test could  
14 easily be done accordingly.

15 **DR. DANIEL SCHLENK:** Okay. Dr. Pettis?

16 **DR. JEFF PETTIS:** Just a bit of followup of  
17 when Jens said that in doing these cage studies, you often see  
18 that they use random age bees and for a longer test, it would  
19 not be appropriate. You would want to start the test with newly  
20 emerged bees.

21 **DR. DANIEL SCHLENK:** And Dr. Ostiguy?

22 **DR. NANCY OSTIGUY:** I concur with what's been  
23 said.

24 **DR. DANIEL SCHLENK:** Okay. Any other Panel  
25 members? Okay. Mr. DeCant, any questions of clarification?  
26 Okay. Let's move on to C please and read that into the record.



1                   **MR. JOSEPH DECANT:** Joe DeCant, EFED, question  
2 10, part C - although a 10-day adult and seven-day larval  
3 toxicity tests have been proposed, please comment on whether  
4 alternative durations of pesticide exposure may be more  
5 appropriate for determining chronic toxicity for adult and  
6 larval bees at a Tier I screen.

7                   **DR. DANIEL SCHLENK:** And Dr. Berenbaum?

8                   **DR. MAY BERENBAUM:** A 10-day test of adult  
9 survival can be informative, but interpreting the biological  
10 meaning outside the social context remains challenging. Rather  
11 than an alternative duration of exposure, an alternative design,  
12 which we've all ready discussed, might be considered. Survival  
13 could be assessed in social context with the use of these cage  
14 brood or broodless bee cup tests, which have been used with some  
15 success in other contexts.

16                   As for the larval toxicity tests as we stated  
17 earlier, estimating the test duration through pupation rather  
18 than terminating it at an arbitrating interval would be most  
19 appropriate.

20                   **DR. DANIEL SCHLENK:** Dr. James?

21                   **DR. ROSALIND JAMES:** I concur with what was  
22 said. Just to clarify because she said it quickly, I mean there  
23 is an alternative way of doing brood tests. The standard tests  
24 are putting brood inside a container like a 48 well plate and  
25 feeding them by hand and cleaning them, but you can also cage  
26 adult bees with some brood and allow the adult bees to care for



1 the larvae, and you can still monitor the mortality of the  
2 larvae.

3 **DR. DANIEL SCHLENK:** Dr. Pettis?

4 **DR. JEFF PETTIS:** I concur with what's been  
5 said.

6 **DR. DANIEL SCHLENK:** And Dr. Ostiguy?

7 **DR. NANCY OSTIGUY:** I concur.

8 **DR. DANIEL SCHLENK:** Any other comments? Mr.  
9 Pistorius?

10 **MR. JENS PISTORIUS:** Thank you. Maybe chronic  
11 toxicity is a question of definition for the larva bees. It is  
12 for the larval test always the question of dosing. Is it acute  
13 dosing at one day of feeding or dosing on every day of feeding?  
14 We discussed this in the group and we were of the opinion that  
15 everyday should be - the pesticide should be added to food every  
16 day.

17 **DR. DANIEL SCHLENK:** Other Panel members?  
18 Yeah, Dr. James?

19 **DR. ROSALIND JAMES:** I'll just add to that a  
20 little bit. Essentially the larval feeding period is so short.  
21 A difference between acute and chronic, you know, it's hard. We  
22 have difficulty defining it. It's such a short period. It  
23 should be treated, perhaps like chronic, but it is still not a  
24 very long test.

25 **DR. DANIEL SCHLENK:** Dr. Tarpy?

26 **DR. DAVID TARPY:** This is Dave Tarpy. I



1 think, to Dr. James's point, that that's why we avoided in  
2 question 8 beginning this morning, we're seeing a one-time  
3 application versus a continuous application type of language  
4 rather than acute and chronic, just to avoid that distinction of  
5 previous usage of that.

6 **DR. DANIEL SCHLENK:** Anyone else? Dr. Hunt?

7  
8 **DR. GREG HUNT:** Well, I'll just reiterate what  
9 has been said before. The reason behind this is that it's more  
10 biologically relevant for the larvae because they're expected to  
11 be exposed to chronic doses.

12 **DR. DANIEL SCHLENK:** Okay. Dr. Klaine?

13 **DR. STEPHEN KLAINE:** Yeah, but I don't want to  
14 lose something that Dr. Delclos mentioned earlier, and that is  
15 timing of pesticide application. Really, the results are not  
16 really known. So, while for this particular test and a  
17 standardized test, it might be worthwhile to have pesticide dose  
18 to each day. Somebody needs to look at what happens when they  
19 only get in on, say day three or on day five, in terms of the  
20 development and ultimately the affect on things like pupation,  
21 et cetera.

22 **DR. DANIEL SCHLENK:** Dr. James?

23 **DR. ROSALIND JAMES:** But within the colony,  
24 you're going to have all different life stages going on at the  
25 same time, so within a colony - my name is Rosalind James -  
26 within a colony, you're going to have all the different life



1 stages going on at once, and you'll also have all this  
2 trophallaxis and storage. It's just not - it's really rare that  
3 they're just going to one day get a dose in a colony and then  
4 it's all gone.

5 **DR. STEPHEN KLAIN:** Steve Klaine - I agree  
6 with you about the different life stages going on at the same  
7 time, but in terms of from an exposure situation, it's not  
8 unrealistic to think that they would have a pulsed exposure  
9 where they might only see the pesticide one day or two days,  
10 depending on how that exposure occurs.

11 **DR. DANIEL SCHLENK:** Dr. Berenbaum?

12 **DR. MAY BERENBAUM:** Physiologically, it would  
13 be very useful and informative to know when pesticides exert  
14 their effects throughout the development, of course, their  
15 development of larvae. But in terms of the ecology of exposure,  
16 it's difficult to imagine how larvae would be subjected to acute  
17 exposures that weren't also at the same time horrifically  
18 catastrophic. So, you can imagine some kind of direct spray to  
19 the entire colony. But, larvae don't go anywhere and everything  
20 they eat is handed to them and it's already processed food. So,  
21 it's already been through several levels of - well it's been  
22 through several layers of bees actually.

23 So, my bee biology colleagues, correct me if  
24 I'm wrong, but it's hard to conceive of a scenario of acute  
25 exposures that recur frequently at different developmental  
26 stages.



1                   **DR. DANIEL SCHLENK:** Dr. Ostiguy?

2                   **DR. NANCY OSTIGUY:** I think we might be  
3 talking about two different things here. Dr. Klaine, I think  
4 you're correct that it's a data gap that we don't know at what  
5 point we might have physiological changes that trigger in terms  
6 of Tier I or Tier II. I don't know that we need that kind of  
7 test done by an applicant. So, it's a data gap in terms of  
8 understanding some of the information that we might get from a  
9 Tier I or Tier II test, but it's not probably necessary to do  
10 during a Tier I or Tier II test.

11                   **DR. STEPHEN KLAINE:** This is Steve Klaine.  
12 You interpreted me correctly. Thanks.

13                   **DR. DANIEL SCHLENK:** Dr. Pettis?

14                   **DR. JEFF PETTIS:** I agree with Dr. Berenbaum  
15 that the majority of the food is processed and goes into larval  
16 development, but there is some direct pollen feeding and direct  
17 honey that is fed in late larval stage. So, there could be  
18 scenarios where you might see things coming into the hive that  
19 are then used directly.

20                   **DR. DANIEL SCHLENK:** It seems to me that  
21 you're kind of interested in windows of susceptibility during  
22 that developmental stage. So, when are those sort of critical  
23 periods of susceptibility that, you know, changes maybe an  
24 outcome that happens at that day, but wouldn't happen in another  
25 exposure period. I think that's - yeah. Any other comments or  
26 discussion? Okay. Let's move onto letter D. Sorry, I didn't



1 ask for clarification questions, I'm sorry. Mr. DeCant?

2 **MR. JOSEPH DECANT:** Yeah, Joe DeCant, EFED.

3 Earlier in the discussion, Dr. James, I think you brought up  
4 the caged bee test, and that came up earlier in some of the  
5 Panels' comments. Is it a semi-field study that you are talking  
6 about or is this a laboratory based?

7 **DR. DANIEL SCHLENK:** Yes, Dr. Tarpy?

8 **DR. DAVID TARPY:** This is Dave Tarpy, NC  
9 State. These are in vitro studies, so they're all done in an  
10 incubator under controlled conditions, so these would be Tier I.  
11 However, I think what was just mentioned is by combining adults  
12 and larvae in vitro starts to blur Tier I and Tier II, but there  
13 are Tier I group bioassays that have been used successfully.  
14 That's what mostly we've been referring to up until now.

15 **DR. DANIEL SCHLENK:** Yes, Dr. James?

16 **DR. ROSALIND JAMES:** We had some discussions  
17 about how you would do the dosing. I mean, perhaps you could  
18 dose the larvae directly by hand and then have the adult bees  
19 take care of them so the exposure of the adult bees was minimal  
20 and you could make it a larval test. But, you're using a little  
21 more natural method for caring for the brood. When you do it  
22 yourself by hand, you maybe feed the bees every four hours or  
23 how often you feed them, and you have to clean out the wells.  
24 It's very labor intensive to do honey bee larval bioassays.  
25 It's a lot easier to let the nurse bees do this for you and they  
26 feed about every half hour, so the brood gets better care, and



1 it would just be a little natural setting and this was one idea.

2 So, it would be easier to conduct, I think, and the brood would  
3 get better care, so you would have lower control mortality.

4 **MR. JOSEPH DECANT:** Joe DeCant, EFED. So,  
5 maybe just a clarification that this cage of brood study would  
6 be a replacement for the larval toxicity study to be able to  
7 take it through pupation? Or is it an alternative design that  
8 could be used, but the larval toxicity study as we have it in  
9 the Tier I screen would be an appropriate replacement given that  
10 we can take it through pupation. Once it goes through pupation,  
11 it would be sufficient in lieu of the cage brood study.

12 **DR. DANIEL SCHLENK:** Dr. James?

13 **DR. ROSALIND JAMES:** Sorry, Dr. Tarpy, are  
14 there standard methods published on this method? I am familiar  
15 with the method, but I'm not familiar with the literature on it.

16  
17 **DR. DAVID TARPY:** This is Dave Tarpy. I don't  
18 think there are standard methods on a caged brood study, but it  
19 is certainly, I think, feasible. But I don't think there is a  
20 standard think like the Aupinel paper on the hand rearing of  
21 larvae.

22 **DR. DANIEL SCHLENK:** Yes, Dr. James?

23 **DR. ROSALIND JAMES:** So, in lieu of that  
24 answer, my answer to you would be it would be an alternative  
25 method that could be used.

26 **DR. DANIEL SCHLENK:** Dr. Ostiguy, did you have



1 something to add? No. Dr. Berenbaum?

2 **DR. MAY BERENBAUM:** And Aupinel offers the  
3 benefit of harmonization with European ongoing efforts. There  
4 are advantages to that, at least until method alternatives are  
5 developed.

6 **DR. DANIEL SCHLENK:** Okay. Any other  
7 questions Mr. DeCant? Okay. Let's move onto D.

8 **MR. JOSEPH DECANT:** Joe Decant, EFED, question  
9 10, subpart (d) - the White Paper identifies NOAEC as the  
10 chronic toxicity measurement endpoint. Please comment on the  
11 possible use of ECx values as a measure of chronic toxicity for  
12 use in RQ calculations.

13 **DR. DANIEL SCHLENK:** Dr. Berenbaum?

14 **DR. MAY BERENBAUM:** I must confess, there was  
15 a considerable amount of confusion about this question. If I'm  
16 not answer the question you're asking, I'm hoping that associate  
17 discussants will address it better.

18 But the gist of our conversation generally is  
19 that the proposed regression based method for calculating ECx  
20 levels seems feasible if a sufficient number of concentrations  
21 can be tested to allow for confident estimates given the  
22 competence levels do drop at the high and low ends of dose  
23 response curves.

24 Some discussion ensued on including a temporal  
25 component of chronic effects to provide a quantitative estimate,  
26 sort of a lethal time estimate like quantitative analysis of



1 survivorship curves using Kaplan-Meier type survivorship as a  
2 way of estimating chronic impacts. I will leave it to the  
3 associate discussants to elaborate.

4 **DR. DANIEL SCHLENK:** Dr. James?

5 **DR. ROSALIND JAMES:** I like the use of  
6 something like a survivorship analysis that gives you a  
7 different kind of endpoint, but it doesn't give you and  
8 effective concentration because it gives you a time to effect  
9 for a given dose, and then has to be repeated at different  
10 doses. So, the calculation of the RQ wouldn't fit very well in  
11 the survivorship type of analysis. I don't have a better answer  
12 for you than that.

13 **DR. DANIEL SCHLENK:** Dr. Ostiguy?

14 **DR. NANCY OSTIGUY:** I concur with what's been  
15 said.

16 **DR. DANIEL SCHLENK:** Dr. Pettis?

17 **DR. JEFF PETTIS:** I have nothing to add.

18 **DR. DANIEL SCHLENK:** Okay. Other Panel  
19 members? Yes, Dr. Fefferman?

20 **DR. NINA FEFFERMAN:** So, I don't know if any  
21 of them would wind up appropriate, but there are a diversity of  
22 different measures for combining dose response and survival  
23 curves from the epidemiological human literature. So, that  
24 might be somewhere to look for methods that might fit with your  
25 requirements somewhat well, specifically in both cancer research  
26 for carcinogens, but also more specifically even in cancer for



1       teratogens. The teratogen studies and time-tell spontaneous  
2       abortion or miscarriage is a place where I've seen a diversity  
3       of measures that might work really well for you.

4               **DR. DANIEL SCHLENK:** Any other input? Okay.  
5       Oh, sorry, Mr. Pistorius?

6               **MR. JENS PISTORIUS:** Maybe just a comment - in  
7       the coming OECD Guidance document, there is an attempt to  
8       integrate this chronic toxicity measurement endpoints and using  
9       ECx values and the use for risk assessment in trigger values.

10              **DR. DANIEL SCHLENK:** Okay. If we could maybe  
11       reference that in the minutes, that would be good. Okay. Any  
12       other comments from the Panel on D? Okay. Mr. DeCant, any  
13       questions of clarification or Ms. Garber? Yeah?

14              **MS. KRIS GARBER:** Yes please, Kris Garber. I  
15       think it might be a little helpful just to clarify the question  
16       a little bit, but I don't think we're quite getting what we're  
17       looking for, would that be all right?

18              **DR. DANIEL SCHLENK:** Sure, yeah.

19              **MS. KRIS GARBER:** Okay. So, just to put it in  
20       my own words, you know the endpoint that we traditionally rely  
21       upon to measure a chronic duration of exposure would be the no  
22       observed adverse effects level and you know, the design of that  
23       study. The study is designed in a particular way to derive that  
24       NOAEC concentration and, you know, there's been a lot of  
25       discussion in the scientific community about perhaps using an  
26       ECx value, and X would be whatever we decided to be biologically



1 relevant. You see 10, you see 20 or EC50, of course, is  
2 relevant as well, and that would have, you know, a different  
3 study design as well.

4 So, you know the specific endpoint that we  
5 would be going after for a chronic study is really relevant for  
6 considering the design of the chronic study. So, we're just  
7 curious on some thoughts for, you know, looking at effects to  
8 bees and the endpoints that you have all been discussion of the  
9 strengths and limitations of the NOAEC versus the EC whatever X  
10 is. I hope that helps.

11 **DR. DANIEL SCHLENK:** Dr. Berenbaum?

12 **DR. MAY BERENBAUM:** We, or at least I was a  
13 little confused about this question and how it relates to the  
14 next question, which seems to be setting the EC level, question  
15 C. So, maybe we can -- what percent effect - should I go on to  
16 C? E, right, sorry. I am myopic. I just wondered if we could  
17 meld these two, because what you're asking is where should EC be  
18 set and should we use EC? No.

19 **DR. DANIEL SCHLENK:** No, she is not asking. I  
20 believe she's asking would you rather use a NOAEC or an ECx  
21 value. And which one, what are the benefits and you know, cost  
22 associated with those different measures essentially? Because  
23 one is a more deterministic sort of endpoint and the other one  
24 is much more based on the probabilistic curve. You set a value,  
25 which could be 5 percentile, 10 percentile, 20 percentile. I  
26 don't think she is asking what X should be, I think she's asking



1 which method do you want to use. Would it be better to use, you  
2 know, a single point that's statistically significant above the  
3 note in terms of a single point value that you have in your data  
4 set or a derived value that you get from a probabilistic curve.  
5 That's what it sounds like. Yes, Mr. Pistorius?

6 **MR. JENS PISTORIUS:** I hope this is slightly  
7 an answer or maybe helps. I assume now that for the chronic  
8 toxicity testing to determine the NOAEC, you would use the  
9 10-day tests. And then, it is the question how do you actually  
10 come closer to the dosing that you had testing. Do you go from  
11 an acute oral LD50 test identifying doses where you have no  
12 LD50? Then it will be a matter that you have to test quite a  
13 large number of doses to get a reliable level. Also, usually we  
14 know that with no-observed effect of concentrations and a low  
15 observed, that's why for other tests always the LD50 is used.  
16 The LD50 gives a lot higher certainty. So, that may be an  
17 argument to rather use the ECx values.

18 **DR. DANIEL SCHLENK:** Yes, Dr. James?

19 **DR. ROSALIND JAMES:** My brain is hurting on  
20 this question. But, the no-effect level will be more  
21 conservative, and maybe with some of these complex behavioral  
22 things, we want to trigger up higher tiered level. So, that  
23 would be an argument for the no-effect level. But, I don't know  
24 that we really know enough about the statistical variability  
25 that we're going to get out of these bioassays to really answer  
26 that question in terms what you're saying, unless you have



1 experience with that.

2 I really don't know how much variability  
3 you're going to have. Because sometimes like, you know, an EC10  
4 or an EC20 level may have a high amount of variability  
5 associated with it. It might be easier to draw a line at the  
6 no-effect level. You know, we are not always looking for the  
7 easy way out, but in some ways, triggering the Tier II for the  
8 behavioral effects might be a good thing to do if some effect is  
9 seen.

10 **DR. DANIEL SCHLENK:** Dr. Berenbaum?

11 **DR. MAY BERENBAUM:** The one advantage of EC  
12 versus no-adverse effect level is that it's always an open  
13 question just to why no effects have been observed. So, an EC  
14 measurement at least is quantifying a known response to the  
15 degree that that's reassuring and repeatable that could be an  
16 advantage for using EC rather than no observable effect.

17 **DR. DANIEL SCHLENK:** Dr. Klaine?

18 **DR. STEPHEN KLAINE:** Yeah, let me weight on  
19 this. As you indicated before, using the NOAEC has a problem in  
20 that fact that it's heavily dependent on the on experimental  
21 design. So, I'm not sure that the issue with bees has any  
22 relevance to which one you use. I think that the better choice  
23 would be the EC number because its derived in a more realistic  
24 or more probabilistic manner. But, I'm really pleased that you  
25 brought up the issue of variability because, Dr. James, because  
26 I think that's the real driver here. We all use EC50s because



1 it's the most accurate. However, if you have the ability to use  
2 a higher end, you then drive the uncertainty regarding an EC5.  
3 You decrease that uncertainty. So, it may make it more  
4 realistic with this organism to actually generate EC5s than with  
5 classic toxicological organisms where it's impossible to get the  
6 N above the 3 or 4 or 5. So.

7 **DR. DANIEL SCHLENK:** Mr. Pistorius?

8 **MR. JENS PISTORIUS:** I think that in any case,  
9 it is an old problem that both the ECx values and the NOAEC  
10 levels are reported from those studies. So, I just question  
11 which one to use later on, but you can always, from those  
12 studies, get both values.

13 **DR. DANIEL SCHLENK:** Dr. Ostiguy?

14 **DR. NANCY OSTIGUY:** If there is a question  
15 about a low sample size, then my preference is the NOAEC. But  
16 for a large sample size, setting a low EC value, that would  
17 absolutely be preferable. So, the larger sample size would  
18 certainly produce better data.

19 **DR. DANIEL SCHLENK:** Any other comments? Ms.  
20 Garber, does that answer your question? Oh, Mr. Sappington?

21 **MR. KEITH SAPPINGTON:** Keith Sappington. I  
22 think, yeah, this question is difficult. The NOAEC versus the  
23 hypothesis testing, i.e. NOAEC versus point estimation ECx issue  
24 has been around for decades. So, that issue is generic. I  
25 think it would be helpful - and there are advantages and  
26 disadvantages to each one, and there have been a number of



1 papers published on that.

2 In my mind, it would be helpful to know if  
3 given the biology of the honey bee or non-Apis bees, but given  
4 the social nature of at least honey bees, might there be an  
5 Apis-specific reason for favoring one over the other. So, that  
6 is kind of perhaps where we're coming from, because this is an  
7 issue for all tox tests.

8 **DR. DANIEL SCHLENK:** Anybody want to comment  
9 on that? Yes, Dr. Hunt?

10 **DR. GREG HUNT:** I don't know much about  
11 testing pesticides, but you can expect with honey bees to have  
12 with this kinds of tests, kind of small sample size and hive  
13 variability. So, that should factor into your thinking.

14 **DR. DANIEL SCHLENK:** Okay. Dr. Berenbaum,  
15 since you're kind of the lead discussant, do you need any more  
16 input in terms of how to answer that? Or are you good?

17 **DR. MAY BERENBAUM:** I don't think so.

18 **DR. DANIEL SCHLENK:** Okay. Dr. Klaine?

19 **DR. STEPHEN KLAINE:** Not to beat this, but Dr.  
20 Hunt, why do we have to have small sample size?

21 **DR. GREG HUNT:** Well, you don't have to have  
22 small sample size, of course, given enough resources, but it's  
23 difficult to get a lot of incubators and a lot of cages and to  
24 the practicality of finding emerging brood, which you need to  
25 emerge in an incubator and collect the bees, get it all going at  
26 the same time before, you know, they start dying from lack of



1 food and there are practical constraints on it. And we see in  
2 cage studies hive variability without treatment in terms of  
3 mortality.

4 **DR. DANIEL SCHLENK:** Mr. Pistorius?

5 **MR. JENS PISTORIUS:** One remark, I just saw  
6 that in the OECD Guidance document, which would be out for  
7 public commenting next week, I think, there is an approach  
8 which, and also a small discussion about which could be used and  
9 why. So, just as a recommendation maybe.

10 **DR. DANIEL SCHLENK:** We will include that  
11 reference as a document to come, I guess, as you meant.

12 **MR. JENS PISTORIUS:** It will be out for public  
13 consultation, so everybody can vote on this by the end of next  
14 week as a visual document.

15 **DR. DANIEL SCHLENK:** Great. Okay. You have  
16 what you need now? Let's move on into letter E.

17 **MR. JOSEPH DECANT:** Joe DeCant, question 10,  
18 subpart (e) - please provide comments on what percent effect  
19 would be considered a relevant measure of chronic toxicity for  
20 individual bees given the potential compensatory effects which  
21 honey bee colonies may exhibit relative to the effects of  
22 pesticides.

23 **DR. DANIEL SCHLENK:** And Dr. Berenbaum?

24 **DR. MAY BERENBAUM:** Compensatory responses are  
25 essentially a society-level phenomenon, so, inherently or maybe  
26 better suited for Tier II assessment. Recruitment of precocious



1 foragers provides a short-term solution to colonies but may  
2 exert long-term costs on colony health. There is one study  
3 cited in the White Paper, Khoury et al. 2011, that estimates a  
4 reduction of worker longevity of 2.8 days corresponds to a loss  
5 of 0.35 bees per day.

6 With that said, then aiming for an EC level  
7 that takes that number into account may be an approach for  
8 addressing the compensatory behavior. How that translates,  
9 we're not sure, but the recommendation is to be conservative.  
10 So, if it's not 35 percent, because its 0.35 bees per day, but  
11 in general, based on discussions yesterday, it seemed that a  
12 level of EC10 or EC20 might be conservative and attainable under  
13 these experimental conditions.

14 What we think the question is at what point  
15 will worker losses send the colonies into the irretrievable  
16 death spiral. And again, there was a general subjective  
17 impression that 35 percent is not sustainable documenting that  
18 with literature was difficult. So, there is basically one  
19 study, possibly two studies. That was the extent of our  
20 discussion.

21 **DR. DANIEL SCHLENK:** Dr. James?

22 **DR. ROSALIND JAMES:** Maybe some of the  
23 modeling that we'll discuss later today or tomorrow will help  
24 answer this question?

25 **DR. DANIEL SCHLENK:** Dr. Ostiguy?

26 **DR. NANCY OSTIGUY:** I concur with the



1 comments.

2 **DR. DANIEL SCHLENK:** And Dr. Pettis?

3 **DR. JEFF PETTIS:** Just kind of reiterating  
4 what Dr. Berenbaum said that there was a general feeling that  
5 somewhere around 30 percent loss of foragers might be a tipping  
6 point, if you will, on the colony level and then maybe something  
7 slightly more conservative than the 20 percent realm might be  
8 reasonable, but we don't have good data to back that up. And  
9 yes, maybe a modeling approach would help.

10 **DR. DANIEL SCHLENK:** Okay. Any other - yes,  
11 Dr. Hunt?

12 **DR. GREG HUNT:** Just to clarify, and I'm not  
13 sure, but I think that we're talking about like a 30 percent  
14 ongoing loss. Correct me if I'm wrong, because a one-time loss,  
15 they could recover from that.

16 **DR. DANIEL SCHLENK:** So, an ongoing loss of 30  
17 percent sounds like the estimate, right? Dr. Tarpy?

18 **DR. DAVID TARPY:** This is David Tarpy. I  
19 should also note that in a full colony, we're still in Tier I  
20 here, but in a full colony situation that that's very  
21 environmentally contextual, so that different times of year,  
22 colonies are more or less susceptible. So in the fall when  
23 they're populations are naturally declining, a smaller  
24 proportion of bee loss would probably have a much larger impact  
25 versus in the spring when their colonies are rapidly increasing,  
26 presumably more likely to be able to absorb that type of loss.



1 But again, modeling approaches I think would be very informative  
2 here.

3 **DR. DANIEL SCHLENK:** Dr. Fefferman?

4 **DR. NINA FEFFERMAN:** So, just quickly - my  
5 reason for our needs on that if that the ramp up is actually  
6 supporting the continued ramp up, so that if you decrease too  
7 much during that increased growth, what you've done has lost the  
8 capacity to support increased queen fecundity. So, you're  
9 right, I want to model it.

10 **DR. DANIEL SCHLENK:** Mr. Pistorius?

11 **MR. JENS PISTORIUS:** I would just like to  
12 point out that in my opinion it is not appropriate to state 30  
13 percent mortality no matter at what time of the season is  
14 acceptable in this discussion here, and also later on I think,  
15 30 percent mortality even of foragers could be compensable by  
16 the colony, but this may be my perspective as a risk assessor is  
17 not acceptable.

18 **DR. DANIEL SCHLENK:** Yes, Dr. Ostiguy?

19 **DR. NANCY OSTIGUY:** Actually, I think what our  
20 discussion was that if it hit 30 percent, the colony was at the  
21 point of no recovery. So, the level of acceptable loss level  
22 has to be set less than that. The problem is that we don't have  
23 data to say where that should be. Our sense was somewhere  
24 around 20 percent.

25 **DR. DANIEL SCHLENK:** Yes, Mr. Pistorius?

26 **MR. JENS PISTORIUS:** I think that even in some



1 cases, colony can recover if the damage is more than 30 percent  
2 because we've had some incidents where larger percentages were  
3 damaged. But on the other hand, I'm not quite sure if this is  
4 the issue of the questions that we are currently discussing. I  
5 don't know if we're supposed to intend to extend this discussion  
6 at this moment.

7 **DR. DANIEL SCHLENK:** Dr. Ostiguy?

8 **DR. NANCY OSTIGUY:** I think what we were  
9 talking about was a continuous 30 percent loss, not a one-time  
10 loss, because certainly colonies can recover from a significant  
11 loss depending on environmental conditions.

12 **MR. JENS PISTORIUS:** I agree, and I think the  
13 difficulty was that we have just been mingling laboratory issues  
14 and realistic world conditions, basically in the discussions.  
15 But I agree to what Nancy Ostiguy has said.

16 **DR. DANIEL SCHLENK:** Okay. Any other  
17 comments? Okay. Mr. DeCant, do you have what you need for that  
18 question? Okay. All right, let's move on to the next  
19 subsection and read that into the record.

20 **MR. JOSEPH DECANT:** Joe DeCant, EFED, question  
21 10, subpart (f) - although the White Paper identifies some  
22 measurement endpoints for assessing chronic toxicity to  
23 individual bees such as survival, please comment on other  
24 endpoints to consider in chronic toxicity studies which the  
25 Panel believes are important and the associated study design  
26 elements.



1                   **DR. DANIEL SCHLENK:** And Dr. Berenbaum?

2                   **DR. MAY BERENBAUM:** As Dr. Tarpy mentioned in  
3 the response to question 8, other endpoints to consider in  
4 chronic toxicity tests may include behavioral impacts, learning  
5 assays, automated recording of motility and behavioral  
6 interactions and the like, although these haven't been vetted  
7 extensively in the context of chronic toxicity.

8                   Future endpoints could include gene expression  
9 relative to detoxification and/or antioxidant status,  
10 immunocompetence senescence traits and the like. Such surrogate  
11 biomarkers are widely used in human risk assessment. They  
12 haven't been validated for bees, but in many ways, they are more  
13 appropriate given the social nature of the organism. With that  
14 said, we don't have any - at least yesterday, discuss any  
15 guidelines known for identifying biomarkers or validating them.

16  
17                   **DR. DANIEL SCHLENK:** Okay. Dr. James?

18                   **DR. ROSALIND JAMES:** I think we answered this  
19 in 8B when we talked, the similar question was proposed for  
20 acute toxicity. Since we were recommending just one test, we  
21 really just answered the question already in 8B, time to  
22 pupation and larval development time, success of pupation, that  
23 sort of thing.

24                   **DR. DANIEL SCHLENK:** And Dr. Ostiguy?

25                   **DR. NANCY OSTIGUY:** One thing that we might,  
26 that would be a future possibility, there has been work done



1 looking at colony level ability to respond to health threats,  
2 would be one way to describe it. And these are responses to  
3 specifically, pathogens is where the work has been done that  
4 occur at a colony level but not at an individual level. And,  
5 that would then be potentially a future - well hopefully a  
6 future test, but there's a lot of data that needs to be  
7 collected to make that at all viable.

8 **DR. DANIEL SCHLENK:** Sounds like the colony  
9 level effects will be discussed later on, yeah? Okay. And Dr.  
10 Pettis?

11 **DR. JEFF PETTIS:** I concur with what has been  
12 said.

13 **DR. DANIEL SCHLENK:** Dr. Fefferman?

14 **DR. NINA FEFFERMAN:** As one potential  
15 mechanism for sublethal kinds of effects like this one that we  
16 haven't discussed so much is response to temporal task  
17 switching. So, most bees have an age-based polyethism of what  
18 they'll do primarily, but in terms of things like health, not  
19 just health threats, but things like if there's a point source  
20 of heat, the recognition of individual bees to go and act as  
21 their coolant batteries themselves, heat shielding, response to  
22 heat shielding, response to solicitation from nurse bees and the  
23 lack of nursing bees from older bees, switching back to nursing  
24 with that stimulus. Those kinds of task switching behaviors  
25 could be critical over chronic toxicity.

26 So, all of the these questions sort of boil



1 down into my mind into how stoned does the average New Yorker  
2 have to be for the busses to stop running? And that's really  
3 hard to measure locally. So, but I haven't heard anyone talk so  
4 much about the communication and solicitation response aspects  
5 of colony organization. I want to put that out there as one  
6 thing to try and measure as we look at this.

7 **DR. DANIEL SCHLENK:** Mr. Pistorius?

8 **MR. JENS PISTORIUS:** In addition to what Dr.  
9 James has said, on the in vitro studies, I just want to refer  
10 the comment I made before for the acute contact and oral  
11 toxicity studies that in this case. Also the behavioral aspects  
12 should be noted and this might, even if not really, a trigger  
13 can be recommended on what portion of bees we see those effects  
14 that the risk assessments needs to have the flexibility to  
15 trigger other tests on this information of sublethal behavioral  
16 abnormalities, especially because when you think that a chronic  
17 toxicity test is aiming at doses going below the LD50 and its  
18 behavioral abnormalities persist, then it might trigger further  
19 concern. The risk assessment needs an option to go to a higher  
20 tier test or to other tests as needed.

21 **DR. DANIEL SCHLENK:** Okay. Any other input?  
22 Yes, Dr. Potter?

23 **DR. THOMAS POTTER:** Yes, this is Tom Potter.  
24 We had a few side bar discussions here at the last break,  
25 discussing the possibility of using respiration measurements as  
26 index of stress. And, I would be interested to hear some



1 comments from the rest of the Panel on whether or not that would  
2 be a useful approach.

3 **DR. DANIEL SCHLENK:** Anybody want to weigh in  
4 on that one? Dr. James?

5 **DR. ROSALIND JAMES:** This is a little remote,  
6 but my first thought is that there really aren't any  
7 standardized sort of methods for relating that to anything,  
8 except there is a fair amount of work done with respiration  
9 rates and diapause in the dormant solitary bees. The honey bee.

10 **DR. DANIEL SCHLENK:** Can you - do you want to  
11 - can you get that on record please?

12 **DR. NINA FEFFERMAN:** Sorry, Nina Fefferman.  
13 Just that there's a lot on colony level respiration needs and  
14 stress and success of colonies in ants, but I don't know of  
15 anything in bees.

16 **DR. DANIEL SCHLENK:** Yes, Dr. James?

17 **DR. ROSALIND JAMES:** At the colony level, so a  
18 Tier II type of test?

19 **DR. NINA FEFFERMAN:** Actually both individual,  
20 but those tend to be challenged type individual studies where  
21 they take an ant and they either an antimate pheromone signal  
22 that signals help, there's a bird or something like that, and  
23 then there's sort of what you would call psychological stress  
24 for individual ants, but then yeah, most of the stuff is really  
25 quite good for regulating stress and colony impact at the Tier  
26 II level, yeah.



1                   **DR. DANIEL SCHLENK:** Yes, Dr. Hunt?

2                   **DR. GREG HUNT:** Well, there are studies in  
3 looking at the flight metabolic rate of honey bees, which have  
4 the highest mass flight metabolic rate of anything, even humming  
5 birds. So, I mean that could be investigated, and it being a  
6 physiological test, it could be a good stress test.

7                   **DR. DANIEL SCHLENK:** Yes, Dr. Pettis?

8                   **DR. JEFF PETTIS:** I was going to follow up on  
9 what Greg said that we have these treadmills that you can  
10 actually just glue the bee to and force it to fly and look  
11 energetic, so it might be - it hasn't been fully developed, but  
12 it might be a way to go about it.

13                   **DR. DANIEL SCHLENK:** So, a flight tunnel,  
14 right? Okay. So, it sounds like there are some other endpoints  
15 that are still in development so to speak that could be, you  
16 know, proposed - physiological biomarkers perhaps, if you want  
17 to put it into a category I guess. Okay. Any other comments on  
18 other endpoints? Okay. I think we got barely enough brain left  
19 for one more question before lunch here if we can squeeze in  
20 this last letter here. So, Mr. DeCant, do you want to read that  
21 into.

22                   **MR. JOSEPH DECANT:** Joe DeCant, question 10,  
23 subpart (g), section 4.1.3 of the White Paper discusses the  
24 uncertainties associated with developing risk assessments based  
25 on studies of sublethal effects when sufficient linkages have  
26 not been developed to understand how the sublethal endpoints may



1 be quantitatively related to typical assessment endpoints, that  
2 is growth, impaired survival, and reproduction, at the whole  
3 colony level. Please comment on the proposal to use data on  
4 sublethal endpoints to qualitatively, that is, no Risk Quotients  
5 are derived to characterize effects and risk until sufficient  
6 linkages between these endpoints and the defined assessment  
7 endpoints have been developed as in the Adverse Outcome  
8 Pathways.

9 **DR. DANIEL SCHLENK:** Dr. Berenbaum?

10 **DR. MAY BERENBAUM:** The social environment  
11 complicates the process of identifying reliable, reproducible  
12 and relevant sublethal endpoints. But given the complexities,  
13 sublethal endpoint use is conservative and is very important for  
14 providing insight into super-organism responses and they are  
15 critical triggers for signaling a need for moving onto Tier II  
16 or higher tiers. So, we would be considered generally advisable  
17 not to discount or dismiss sublethal endpoints.

18 An important priority for future research  
19 though, would be developing assessments for neurotoxicity  
20 symptoms. Although direct links to colony health may not yet be  
21 fully understood, they're indicative of biological activity of  
22 chemicals and therefore appropriate candidates for further  
23 investigation at the colony level.

24 So, the results of these types of studies can  
25 demonstrate a need for additional quantitative tests at Tier II.  
26 Eventually, links have to be made, but given the state of



1 knowledge at the present time, sublethal endpoints are really  
2 critical indicators for biological activity that may be  
3 difficult to assess using traditional toxicological measures  
4 designed for organisms that are not so dependent on a social  
5 environment.

6 **DR. DANIEL SCHLENK:** Okay. Dr. James?

7 **DR. ROSALIND JAMES:** I concur, but I will add  
8 in defense of the nonsocial bees, they have complex behaviors  
9 also in nest building and so on. It would be applicable to the  
10 non-Apis bees as well.

11 **DR. DANIEL SCHLENK:** Dr. Ostiguy?

12 **DR. NANCY OSTIGUY:** Just in case this matters,  
13 there is not a 4.1.3, so I think it's a 4.1.2.1. So, we think  
14 we're answering the correct one, but let us know if we're not.

15 **DR. DANIEL SCHLENK:** Okay.

16 **DR. MAY BERENBAUM:** This is May Berenbaum.  
17 4.1.2.1.2 is the section.

18 **DR. DANIEL SCHLENK:** Yeah. I got that one.  
19 Okay. Dr. Pettis?

20 **DR. JEFF PETTIS:** I concur.

21 **DR. DANIEL SCHLENK:** That we're on the right  
22 number?

23 **DR. JEFF PETTIS:** We actually spent quite a  
24 bit of time trying to find the 4.3. Are we missing something,  
25 but no we haven't.

26 **DR. DANIEL SCHLENK:** Just a challenge, it was



1 a challenge. Yes? Dr. Fefferman?

2 **DR. NINA FEFFERMAN:** Sorry, in all  
3 seriousness, this is yet another one that I think we'll end up  
4 discussing the context for later. But really anytime that these  
5 questions relate to how to scale unknown relationships linking  
6 individual level sublethal effects, or lethal effects frankly,  
7 to the colony level compromise, that's a place where modeling  
8 actually may be able to help. Even working backwards toward a  
9 Tier I type study to recommend what kinds of sublethal behaviors  
10 could be used as proximate measures in individuals that would  
11 then scale up to call any compromise, but we'll talk about that  
12 more later.

13 **DR. DANIEL SCHLENK:** Any other comments? So,  
14 it sounds like at least for adverse outcome pathways,  
15 neurotoxicity would be at least one sort of pathway to pursue  
16 it. I'm interested in the Panel's response to the beekeeper  
17 that had the eviscerated males that were lying around. Would  
18 that indicate maybe an endocrine pathway would be another sort  
19 of target that would be something to explore. Yes, Dr. Hunt?

20 **DR. GREG HUNT:** I don't really know why drones  
21 fall dead with their genitalia everted, but I think it has  
22 something to do with general stress. I'll leave it there.

23 **DR. DANIEL SCHLENK:** Yes, Dr. Tarpy?

24 **DR. DAVID TARPY:** This is David Tarpy. This  
25 is pure speculation, but because the kind of default situation  
26 for drones is to turn themselves inside out to evert their



1 genitalia to mate, but it's some of the lower ganglion that  
2 inhibit that, that it actually suggest a potential neural  
3 pathway for that particular report. But, that's again, just  
4 speculation.

5 **DR. DANIEL SCHLENK:** All right. Any other  
6 comments? Mr. DeCant, do you have questions of clarification?

7 **MR. JOSEPH DECANT:** Joe DeCant, EFED. I just  
8 want to sort of make sure that I understood the panel correctly,  
9 that the sublethal endpoints, some of them are still being  
10 developed. The assays are either still in development or we're  
11 still waiting on the quantitative linkage between the sublethal  
12 endpoints and some of the colony level effects. Given that  
13 though, these sublethal endpoints are important and that while  
14 we may not use them quantitatively, the panel would agree that  
15 we should use them qualitatively and perhaps use them to also  
16 trigger higher tier studies.

17 **DR. DANIEL SCHLENK:** I see most heads nodding  
18 on that one. So, it seems like a general agreement. Okay. All  
19 right, I think we're sufficiently toasted. So, let's take a  
20 lunch break and come back at 12:45.

21 (WHEREUPON lunch was taken)

22 All right, if you would please take your  
23 seats. Okay. Let me just go to the Agency. Do you have any  
24 other clarifying questions that you have before we start with  
25 question 11 on any of the previous questions? We're good?  
26 Great. Okay. Let's go ahead and read 11A into the record if



1 it's up there.

2 **MR. JOSEPH DECANT:** Joe DeCant, EFED, question  
3 11, subpart (a) - although colonies are typically confined to  
4 enclosures for Tier II studies and these enclosures can limit  
5 the environmental realism of the study conditions, tunnel  
6 studies provide an opportunity to collect colony-level effects  
7 and potentially exposure information. Please comment on the  
8 relative strengths, that is foraging activity by adult worker  
9 bees is limited to treated crop; trophyllaxis enabled and  
10 limitations such as limited study duration, smaller colony sizes  
11 and reduced forage area of these methods.

12 **DR. DANIEL SCHLENK:** Lead discussant for  
13 that's Dr. Tarpy.

14 **DR. DAVID TARPY:** This is David Tarpy, NC  
15 State, good afternoon everybody. I want to preface the comments  
16 in talking about Tier II and other studies by kind of briefly  
17 reiterating the point of linking this to our discussion this  
18 morning about Tier I bioassays, and how when we're looking at  
19 social insects such as honey bees that it starts to blur the  
20 lines very much. So, that simple LD50 mortality measurements of  
21 individual adult worker honey bees doesn't actually really  
22 capture the myriad of different exposure pathways and other  
23 effects of social insects. So, this to me is really the  
24 fundamental question that faces the panel and is driving a lot  
25 of our discussions for this White Paper.

26 So, one means of addressing that obviously is



1 to incorporate a lot of these other bioassays that we discussed  
2 in question 8 and others to help bridge this gap, but it still  
3 doesn't really fully serve as a proxy for colony-level  
4 phenotype. And again, this is because honey bee colonies are  
5 like super organisms. So, in that way, biologically the basal  
6 unit is that entire colony. It's very hard to test these  
7 systems in a reductionistic approach.

8 The kind of analogous or an analogy to this is  
9 to study individual cell cultures of say a rat and looking at  
10 LD50s of individual cells of that cell culture and then trying  
11 to extrapolate that to the entire rat. So, the way that it's  
12 actually done is to study individual rats and looking at LD50s  
13 of different compounds of whole rats. This is the same that is  
14 true for entire colonies.

15 So, in some ways, it's inescapable to have to  
16 test entire colonies to find full phenotypic effects at that  
17 level. Not to say that Tier I screens of individual bees or  
18 brood are not informative, it's just that they are not perfect  
19 proxies or surrogates for higher levels at the colony level.

20 So with all that said and with all that in  
21 mind, the question asked to identify some strengths and  
22 weaknesses of Tier II studies, and obviously some to strengths  
23 or because it's measuring at in vivo in this super organism  
24 context, it does better enable and capture a lot of those  
25 complex social interactions that are going on that result in  
26 full fledge colony phenotype.



1           The limitations, many of which are kind of  
2 logistical in that there is difficulty in getting large sample  
3 size when you're dealing with entire colonies. It's just a  
4 realistic problem with trying to get sufficient number of  
5 colonies, especially because they're so highly variable social  
6 insects. One of their keys to their ecological success is their  
7 huge variability and their ability to adapt different  
8 environments. So, there's a lot of variation across colonies.  
9 So, trying to account for that requires higher sample sizes and  
10 that's obviously a real logistical constraint. Again,  
11 automation of data collection and means to be able to streamline  
12 that would be incredibly helpful, but I don't think the  
13 technology is quite there yet.

14           In doing so, it's always helpful - or the  
15 Panels feel that the type Tier II studies, it's very helpful to  
16 contextualize a lot of these colony level phenotypes by always  
17 having adequate control, either positive and/or negative control  
18 groups to relativize it. That helps be able to place the  
19 findings into the proper context. It's hard to have kind of  
20 universal measurements across these different studies because  
21 they're done different times of the year under very different  
22 conditions, environmental conditions. So, doing to contextually  
23 is very helpful.

24           We can discuss this in part B and in other  
25 places, but we do also want to throw out the notion that the two  
26 types of type II studies that were discussed in the White Paper,



1 one was enclosed foraging arenas in cages and then others where  
2 they were on a treated target crop and other where they were  
3 free flying, but they were being actively fed through  
4 supplemental feed these different compounds in question that I  
5 think that the former approach might be more amenable to other  
6 social insects, particularly bumble bees.

7 Honey bees are notoriously difficult to keep  
8 in small enclosed areas. Their foragers always want to be  
9 trying to go outwards, further outwards, and that's why honey  
10 bees are really not used in greenhouse pollination, for example.

11 They're just not as amenable to that type of condition. Bumble  
12 bees, however, which are primitively used social and not highly  
13 used social, but they still live in colonies. They tend to be  
14 much more amenable to those types of systems.

15 For the free-flying type of approach however,  
16 honey bee colonies do have some strengths and would probably be  
17 more amenable to that type of Tier II approach.

18 **DR. DANIEL SCHLENK:** Okay. Thanks. Dr.  
19 Ostiguy?

20 **DR. NANCY OSTIGUY:** Our concern about Tier I  
21 studies that Dr. Tarpy was mentioning because of honey bees  
22 being a social insect may be partially addressed when we talk  
23 about question 14 and I'm going to probably use the wrong word,  
24 Nina, but meta-models. So, the higher level of modeling so we  
25 can select which of the Tier I studies might actually provide us  
26 with the best information about what's going to happen then at



1 the colony level.

2 So, there may be a way for us to address that  
3 social insect issue that in some ways is better studied  
4 empirically or with hard data with Tier II and Tier III, Tier I  
5 still may provide us with some useful information once we have a  
6 better idea of which of those Tier I pieces of data that we  
7 might collect, actually would trigger colony-level problems.

8 **DR. DANIEL SCHLENK:** Thanks. And Dr. Pettis?

9  
10 **DR. JEFF PETTIS:** Just a bit to add. There's  
11 a question about colony size in some of these Tier II and Tier  
12 III studies and how small colonies might not be representative  
13 of large colonies. I think if small colonies are properly set  
14 up, even if their nucleus colonies, they can be somewhat  
15 representative of whole colonies. You actually might - using  
16 small colonies, you actually might see an effect, which you  
17 wouldn't see in larger colonies. So, I just wanted to get that  
18 point in. I concur with the other comments.

19 **DR. DANIEL SCHLENK:** Other Panelists? Yes,  
20 Dr. Berenbaum?

21 **DR. MAY BERENBAUM:** Just one caveat about  
22 supplemental feeding, there are among these types of studies  
23 posed in the literature, there is supplemental feeding and  
24 there's force feeding, and it's important to emphasize that  
25 honey bee behavior is incredibly sophisticated, particularly  
26 with respect to food selection and food evaluation. So, suffice



1 to say that not all supplemental feeding studies may be  
2 equivalently useful.

3 **DR. DANIEL SCHLENK:** Okay. Mr. Pistorius?

4 **MR. JENS PISTORIUS:** I would like to go into  
5 detail on the usefulness of different semi-field studies  
6 described here, those feeding studies later on. Let me say this  
7 all ready, I think it is absolutely -- it has to be refused to  
8 use such feeding studies. They are - for me personally, they  
9 are intermediate between Tier I and Tier II, but they're not  
10 really a Tier II test. I would give the justification for this  
11 rather on the point D when we look at different studies.

12 So, now for me, I'm talking about the OECD 75  
13 and EPPO 170, which I think are very appropriate to with the  
14 worst case enclosure to assess the acute effects on bees. Of  
15 course, there are some limitations we know in the confinement.  
16 For instance, there may be a natural reduction of the brood  
17 because the bees don't want to breed as much under natural  
18 conditions, that in effect what you can eradicate by appropriate  
19 replications. But I think that, especially the EPPO 170 and  
20 OECD 75 are the ones to go for as a Tier II test.

21 You have acute and you've got oral and contact  
22 exposure combined. What has come out of the comments that we  
23 basically want to have a look at all the different exposure  
24 routes and effect on the bee colony. And small colonies, I  
25 agree with Jeff, may be used to interpret the effects on  
26 colonies to some extent. In some cases, it might be a necessary



1 step to go to Tier III, but it gives you a clear indication,  
2 especially the OECD 75 method, I think, is very reliable. For  
3 one, it builds on the EPPO 170, but specifies the brood  
4 assessment.

5 This in general allows to have really worst  
6 case exposure of bees, of different castes of bees. You have  
7 some certain limitations that you have to supply some food, but  
8 that is always the case with every method except one feeding  
9 method where you could reduce the amount to almost zero, but you  
10 have to apply a little bit of food. Of course, there is a  
11 little uncertainty, but I think in general, they are very  
12 appropriate studies, but only the OECD 75 and the EPPO 170, the  
13 semi-field studies - I will elaborate on that later on - the  
14 semi-field feeding studies that have been mentioned here before  
15 in my opinion are not really appropriate as Tier II.

16 **DR. DANIEL SCHLENK:** Dr. Hunt?

17 **DR. GREG HUNT:** I'm not sure I understand you,  
18 Jens. You're saying that in Tier II, you don't think  
19 supplementary feeding studies have any place?

20 **MR. JENS PISTORIUS:** Thank you, I will  
21 elaborate on this. I think three different types of studies  
22 have been mentioned. Please correct me if I'm wrong. First,  
23 the Oomen method. The second one is free-flying colony study,  
24 so it's a field, but still with supplementary feeding. Then you  
25 can do sugar feeding or pollen package feeding. I think this is  
26 not appropriate for Tier II tests.



1 I think the most appropriate is to have a crop  
2 that offers nectar and pollen to over spray bees. During  
3 flowering, you get contact and oral exposure. With the colony  
4 that you feed either sugar syrup or pollen, you'll definitely  
5 have bees that go somewhere else and you will not be able to  
6 quantify the exposure.

7 You will have a portion of bees even if you  
8 can say, okay the sugar water has been consumed and pollen has  
9 been eaten to this extent would never be able to quantify what  
10 exact amount has been. And it's not a worst case exposure  
11 anymore, which is the aim of a Tier II test in my opinion.

12 So, with those feeding methods, I'm not happy  
13 at all. Once exception - the Oomen test can be used, but it's  
14 regarded as intermediate between Tier I and Tier II. The big  
15 advantage, why I still like it a little bit, is that you can  
16 actually do such a dose response. The test method, as it is  
17 described in the literature in 1992 is not appropriate. It  
18 needs modifications. For instance, there is a dosing of one  
19 liter sugar solution, a unique dosing. And this is not  
20 appropriate. It should be dosing over a longer time with syrup,  
21 at least, whatever at least nine or ten days that they get  
22 everyday sugar that you actually - and the big advantage is you  
23 can find some dose response via this.

24 With the other ones, I think you are really  
25 neglecting something. If you do a feeding method, you cannot  
26 quantify exposure and there is a lot of worst case exposure.



1                   **DR. DANIEL SCHLENK:** Dr. Hunt?

2                   **DR. GREG HUNT:** I guess that clarifies it  
3 because you're not talking about tunnel experiments. In a  
4 tunnel experiment, you could quantify exposure. We just set one  
5 up last week. If you think that the exposure will be through  
6 pollen, then you establish colonies with depleted pollen and you  
7 feed them paddies and you measure what they eat.

8                   **DR. DANIEL SCHLENK:** Okay. Mr. Pistorius?

9                   **MR. JENS PISTORIUS:** When I get a study, I'm  
10 happy when I get a study that gives me a lot of information and  
11 not only partial information. What I would suggest to use EPPO  
12 170 and OECD 75 instead of such feeding studies. The reason for  
13 this is that you have a combined exposure. You will not be able  
14 to get a contact exposure of the bees.

15                   With a Tier I, we look at what is the toxicity  
16 for oral exposure, what is the oral toxicity, contact toxicity,  
17 but by making a spray application on the flowering crop where  
18 the bees are actively foraging. This is the only possibility  
19 where you have the option to get both effects of oral and of  
20 contact exposure in one goal. With this feeding study, if you  
21 identified a risk only for oral exposure, okay. But trying to  
22 get that we want to have the worst case situation, which I  
23 believe should be still the Tier II test, then I think those  
24 methods of feeding are not really giving us the worst case.

25                   **DR. DANIEL SCHLENK:** Dr. Tarpy?

26                   **DR. DAVID TARPY:** This is Dave Tarpy. So



1 then, from what I'm hearing then is that that's what you're  
2 arguing that a full-field rather than a semi-field assessment is  
3 best, which I'm not disagreeing with, but what we're talking  
4 about here are Tier II semi-field tests. So, they're not  
5 supposed to recapitulate all of nature, you're supposed to be an  
6 intermediate right? So, while I agree that by forces feeding or  
7 by supplemental feeding to kinda of impose exposure may not  
8 always mimic the proper exposure routes. I think the question  
9 here is do they still have some utility. So, I don't think the  
10 question is are they mimicking realistic means of exposure, but  
11 do they have some sort of utility in effecting final colony  
12 outcome or final colony phenotype. I would probably say that  
13 they do, but I agree that they do not perfectly mimic how they  
14 would be exposed in the full field, right? But this is not full  
15 field, this is a semi-field Tier II level.

16 **DR. DANIEL SCHLENK:** Mr. Pistorius?

17 **MR. JENS PISTORIUS:** I just want you to  
18 clarify for me a Tier II is always a semi-field intolerance, not  
19 free flying. Free flying in nature, whatever, that for me is a  
20 Tier III test.

21 **DR. DAVID TARPY:** This is David Tarpy again.  
22 While in the White Paper, they articulate and discuss the two  
23 types of Tier II studies. One type is in tunnels, and that's  
24 correct. The other is not confined spatially, right? So,  
25 that's what I was referring to in that.

26 **MR. JENS PISTORIUS:** You are right, and this



1 is exactly what I propose to be changed or what I personally  
2 feel is not appropriate to consider free-flying colonies as Tier  
3 II tests.

4 **DR. DANIEL SCHLENK:** Dr. Pettis?

5 **DR. JEFF PETTIS:** Along this line then, if and  
6 again I think in the White Paper, they do talk about a possible  
7 Tier II with feeding meter, either protein or syrup. If we move  
8 that to a Tier III study, do you still think it's not valid to  
9 at least have some level of exposure where you reduce the stores  
10 in the colony prior to the starting of the test and you start to  
11 provide supplemental syrup and/or protein over time? They are  
12 free flying and I agree -- I know one of the comments is going  
13 to be reduction of incoming pollen is not going to be absolute.  
14 We get dearth periods here in the mid Atlantic of two or three  
15 months where there is virtually no incoming nectar. So, we do  
16 get a period when we can do these tests and not have much  
17 incoming outside resources. So, we are providing the resources,  
18 we've limited the amount at that start, and we do feel that  
19 we're getting exposure. I just think that by controlling the  
20 dose a bit, we are in some cases doing maybe a worse case, I  
21 don't know.

22 **DR. DANIEL SCHLENK:** Mr. Pistorius?

23 **MR. JENS PISTORIUS:** I think it is actually  
24 what you mentioned, you are right. I think for some cases it  
25 can actually be used in the field conditions. For instance, now  
26 let's assume that we have a very highly toxic substance and it's



1 even systemic and you have very much concerns and you want to  
2 have this long-term duration really checked.

3 I think as an additional method after you have  
4 had an exposure to the real crop - I mean for instance, let's  
5 have a look at systemic and you have whatever we have in  
6 Germany, a winter oilseed rape crop, which is attractive and  
7 offering nectar and pollen. Then you say, okay maybe the test  
8 crop didn't flower long enough and in real life, you maybe have  
9 different flowering crops and this is the situation which is  
10 very hard to cover when you conduct a field test. To time the  
11 flowering of different crops, I mean that is maybe reality, but  
12 if you want to design the study, that is not possible.

13 So then, I would agree that you can say, okay  
14 at field realistic concentrations, you can add pollen paddies  
15 and you can also do sugar feeding to extend exposure. To have,  
16 you know, a long-term test and to mimic that the exposure is not  
17 like winter oilseed flowering taking place four weeks, you can  
18 mimic that the exposure would be continuous over five weeks to  
19 get a worst case experiment. So I think, this case, additional  
20 pollen and nectar feeding is appropriate, but I think it's  
21 different study design from how I understood -- maybe I  
22 understood it wrong from how I understood what was described.

23 **DR. DANIEL SCHLENK:** Okay. Dr. Hunt?

24 **DR. GREG HUNT:** I think I better understand  
25 what Dr. Pistorius was taking about. You consider Tier II  
26 tunnel experiments, and I agree that is probably the most



1 practical way to go. You have the most control. It's going to  
2 depend on the crop and what you think is the route of exposure.  
3 Some crops, the bees might not forage on in the tunnel, and in  
4 some cases it may be appropriate to do a feeding experiment.

5 **DR. DANIEL SCHLENK:** Okay. Mr. Pistorius, we  
6 need to get on with this. Go ahead.

7 **MR. JENS PISTORIUS:** Just for crops which are  
8 not attractive to bees, it is a question of systemic seed  
9 treatments for instance. I mean, if it's not attractive to  
10 bees, then the question is would you actually have exposure via  
11 nectar and pollen, which would then, as I understood, would not  
12 be the case. Then if you look at other issues, for instance if  
13 you have some guttation issue or whatever, you can do that.  
14 Maybe then, you can do some additional feeding. That's okay,  
15 maybe. But if you have a target crop where you want your  
16 application on, at least what we do and this target crop is not  
17 attractive to bees and we want to test the impact of a spray  
18 application, we use a surrogate flowering crop, which offers  
19 nectar and pollen because we want to know what the pesticide  
20 does. If it is intended not to be used on or if it's intended  
21 to be used on a not flowering crop, which offers no nectar and  
22 pollen, then this is basically a risk mitigation measure.

23 **DR. DANIEL SCHLENK:** Okay. Any other comments  
24 on this? It sounds like that tunnels everybody is good with,  
25 but there may be some issues relate to the semi-field. Is that  
26 what I'm hearing? Yes? Mr. Pistorius?



1                   **MR. JENS PISTORIUS:** May I? Sorry. It's not  
2 tunnel or semi-field for me. For me, a semi-field is a tunnel  
3 study. It is the question, are they free flying and is  
4 additional syrup fit as a main exposure.

5                   **DR. DANIEL SCHLENK:** So, let's just be sure we  
6 document that very well in the written comments. Dr. Tarpy,  
7 you're responsible for that, I guess. Any other comments on  
8 this particular issue? Okay. Let me go to the agency then.  
9 Any specific questions of clarification? I will give you a  
10 moment or two.

11                   **MR. JOSEPH DECANT:** This is Joe DeCant, EFED.  
12 I think that - so question A refers to the tunnel design and  
13 specifically the tunnel design in question. B is going to touch  
14 on the feeding design that Mr. Pistorius was talking about. I  
15 think that maybe for clarification - so what I heard were the  
16 strengths and the weaknesses of the tunnel design sort of at the  
17 first part of the conversation and the discussion. I think what  
18 would be helpful is if the question is about where the feeding  
19 design fits into the tiered approach, if the Tier II is  
20 semi-field and we're talking about tunnel studies, then is the  
21 feeding design, does it sit in between Tier I and Tier II or  
22 does it sit in between the tunnel and the Tier III full field?  
23 Maybe just some clarification on where that might sit and  
24 getting to the utility of it in subpart (d).

25                   **DR. DANIEL SCHLENK:** Do you want an answer  
26 for that now? Yes? Can we answer that now? Dr. Tarpy?



1                   **DR. DAVID TARPY:** This is David Tarpy. Since  
2 I'm lead, I guess I'm the one in the crosshairs on that. I  
3 don't know if there is a clear answer to that. I think in some  
4 ways, it depends on the experimental design. A lot of these  
5 boundaries are blurred because of the social organism that we're  
6 using here as a model for it. In some cases, I would say that  
7 it lies between the Tier I and the food house design. In  
8 others, I would say it would not be. So, I don't know if there  
9 is a definitive answer to that question, if I'm understanding it  
10 correctly.

11                   **DR. DANIEL SCHLENK:** Yes, Mr. Sappington?

12                   **MR. KEITH SAPPINGTON:** Keith Sappington. With  
13 regards to subpart (a) and as you draft the report, it would be  
14 very helpful to the agencies for any insights that you can  
15 provide on the likelihood of false negatives with tunnel  
16 studies, semi-field tunnel studies as well as false positives,  
17 especially if this may end up being sort of the only study being  
18 considered in Tier II. Thank you.

19                   **DR. DANIEL SCHLENK:** Okay. Yes, Dr. James?

20                   **DR. ROSALIND JAMES:** The tunnel studies I've  
21 done have been with solitary bees, and I assume these problems  
22 are exacerbated with honey bees in that say with bumble bees,  
23 you have to feed them inside a greenhouse even when they're used  
24 for greenhouse pollination. So, the idea of being able to treat  
25 the plant in the tunnel and then expect normal foraging, you're  
26 not really going to get normal foraging because you have to have



1 a really big tunnel. Bees need a lot of plants. There is not  
2 very much nectar and pollen produced per flower. So, they  
3 really need many, many plants and they quickly deplete out a  
4 greenhouse. So your dose may be reduced in a tunnel test.

5 **DR. DANIEL SCHLENK:** Okay. Mr. Pistorius?

6 **MR. JENS PISTORIUS:** I agree, maybe a  
7 different issue with bumble bee colonies, although we have also  
8 done some studies with bumble bee colonies, for instance oilseed  
9 rape or Phacelia tanacetifolia, which also offer good nectar.  
10 At least for the time of flowering, you have good exposure and  
11 you were able to keep the colonies maintain - I mean of course,  
12 you have to have a certain amount of food inside a bee colony,  
13 just at least a few hundred grams of honey and a little bit of  
14 pollen. So if there is a weather change for a few days, they  
15 don't starve to death because that will of course, kill your  
16 study.

17 On the other hand, I think that this is still  
18 appropriate. You don't have normal foraging behavior. I agree  
19 because the tunnels may be of limited or they are always of  
20 limited size, but you can keep them and you can document that by  
21 the income of nectar and pollen by when you do the brood  
22 assessment that actually pollen came in. When you do the  
23 assessments for instance on bee brood, you can actually see at  
24 least they were able to, they were consuming the pollen that  
25 they had before, then they stored new pollen and they were able  
26 to take care of the brood for a while.



1           Then to clarify this again, I think this is a  
2   good way of exposing colonies, but then if you want an extended  
3   exposure over the duration of the flowering crop, you can do  
4   additional feeding. That's what I thought. And what you said  
5   with greenhouse application, that may be true that you have for  
6   instance for tomato pollination, that you will have bumble bees  
7   starving a little bit and that you have to feed something  
8   additionally. Maybe it's a different system. I don't know.  
9   What we do is we register the substance. We register a product  
10   and we have to test that this product is safe, not just special  
11   application. So, what we would go for is we make an application  
12   on a flowering crop that is attractive to bumble bees, like  
13   Phacelia and would use these results to interpret the potential  
14   effects of the product on bumble bee colonies. Then what you  
15   can do to extend this again, you do some feeding.

16           **DR. DANIEL SCHLENK:** Dr. James?

17           **DR. ROSALIND JAMES:** So, it would help if you  
18   used something like the Celia that is a very high nectar and  
19   pollen producing plant instead of the crop. But still, if you  
20   have to start feeding at the end, you're effectively diluting  
21   your dose. Well, you're reducing the dose if you're adding - if  
22   you have to feed supplemental nectar and pollen, they're not  
23   collecting it from a crop that was treated.

24           **MR. JENS PISTORIUS:** This is a very good  
25   point, thank you. When I mean additional feeding is possible  
26   for extending the exposure, of course I mean that you have to



1 use spiked food with realistic field concentrations. Because I  
2 agree if you then feed uncontaminated sugar or pollen paddies,  
3 that is again a reduction of the exposure. This is basically  
4 reducing the certainty of the test. Then you get uncertainty  
5 and we don't know how to make the interpretation.

6 **DR. DANIEL SCHLENK:** Okay. Mr. Sappington?

7 **MR. KEITH SAPPINGTON:** Just one clarification.

8 Mr. Pistorius, when you talk about potentially extending the  
9 study with feeding, is that being considered in the context  
10 within a tunnel or a separate study like a feeding study where  
11 they have free-flying bees.

12 **MR. JENS PISTORIUS:** It would be considered  
13 for after the exposure to the flowering crop, so it's not an  
14 extra study. It is with the same colony. After the exposure  
15 period or after the forage in the tunnel is finished, the  
16 flowers are all gone basically, then what you could do is you  
17 add, have to think about opening up that tents so they are free  
18 flying or maybe you can close, but then you add basically sugar  
19 solution, spiked sugar solution and spiked pollen to extend this  
20 period over the blooming period if you have such a concern that  
21 it is in real life maybe more covering the realistic situation.

22  
23 **DR. DANIEL SCHLENK:** Okay. Any other  
24 questions of clarification from the Agency? Okay. Dr. Tarpy,  
25 you want to - oh sorry, first we need to read in the next  
26 question please.



1                   **MR. JOSEPH DECANT:** Joe Decant, question 11,  
2 subpart (b) - please comment on any other types of colony-level  
3 studies that should be considered as part of Tier II.

4                   **DR. DANIEL SCHLENK:** Dr. Tarpy?

5                   **DR. DAVID TARPY:** This is David Tarpy. So,  
6 the discussants talked about three kind of general alternatives  
7 that may be used, not really, well in Tier II as far as inside  
8 hoop houses or in cage studies. One is to utilize the power of  
9 observation hives, which are glass-walled colonies, glass-walled  
10 hives that enable observations of individual nest mate  
11 behaviors. So, comparable to what we were discussing in  
12 question number 8 this morning about how nest mate interactions  
13 and individual behaviors has relevance to colony level  
14 phenotype.

15                   Recapitulating that in a type II or a Tier II  
16 environment that could possibly augment those same types of  
17 things done in Tier I level may very well be helpful. Again, I  
18 think that the exact behaviors and the manner by which those  
19 behaviors are recorded and analyzed is a data gap, but I think  
20 that it is something that should be considered in these colony  
21 level studies, part of Tier II.

22                   Secondly as was eluded to in previous  
23 questions about the blurring of Tier I and Tier II when it comes  
24 to the colony reproductives. So, queens and drones are both  
25 very, very important. Unlike workers, which may be more  
26 amenable to small groups in vitro, it's very, very difficult to



1 do that with queens and drones. So, Tier II studies ought to  
2 consider and incorporate means by which phenotypic effects on  
3 individual queens and drones can be measured, because those are  
4 in some ways a consequence of colony level phenotype. So  
5 measuring the reproductive potential of queens, the reproductive  
6 potential of drones is something that could also be folded into  
7 this level of assessment.

8 Then finally, there was discussion earlier  
9 this morning about possible Tier II type studies that are done  
10 in vitro, right? So, having small colonies with functional  
11 queens, workers of all ages and brood that they're actively  
12 raising, but done in incubators and not actually in hoop houses  
13 or done outside, but in more controlled environments. These  
14 could probably be done for a period of days, may be weeks  
15 depending on the size and other factors.

16 So these are, what I would consider Tier II  
17 studies because they're involving whole-level colony phenotype,  
18 but they're actually done in vitro. So again, it's this  
19 blurring of these different tiers when it comes to looking at  
20 super organisms.

21 **DR. DANIEL SCHLENK:** Okay. Thank you. Dr.  
22 Ostiguy?

23 **DR. NANCY OSTIGUY:** I concur.

24 **DR. DANIEL SCHLENK:** And Dr. Pettis?

25 **DR. JEFF PETTIS:** I concur with the comments  
26 of Dave. I will just add a bit about some of these potential in



1 vitro kind of cage trials, it would more mimic a whole colony.  
2 We actually did some of those where we took a small - 100  
3 workers and a queen and introduced a known number of eggs to the  
4 colony and asked them to rear them and looked at the success  
5 rate of that small unit to rear that brood. It was pretty  
6 telling. I mean, you can get results with a fairly simple set  
7 up. They're not refined. They're not, in all cases - but I  
8 think that is something that moves between straight Tier I and  
9 Tier II where we begin to capture the super organism.

10 **DR. DANIEL SCHLENK:** Any other panel members?

11 Yes, Dr. Hunt?

12 **DR. GREG HUNT:** Obviously, we're throwing out  
13 a lot of ideas on possible assays, and only a few of these will  
14 probably end up being the ones considered until there is further  
15 evidence of their effectiveness. But one possibility for  
16 another one is to - since there is a general feeling among  
17 beekeepers and bee researchers that perhaps we're seeing more  
18 problems with queens lately. There are indications that certain  
19 compounds like the miticides that beekeepers use in their hives,  
20 the queens are more sensitive to.

21 So, one study that can be done in two weeks'  
22 time is to rear queens in a somewhat contaminated hive. This  
23 can be done very rapidly. The question is what is the best  
24 method to contaminate the colony, and I see Dr. Pistorius is  
25 raising his hand, so he has an idea on this.

26 **DR. DANIEL SCHLENK:** Mr. Pistorius?



1                   **MR. JENS PISTORIUS:** I do have an idea. What  
2 you could do is make a shook swarm, which is basic for the  
3 non-beekeeper that you put the bees without the queen in a cage,  
4 in a box and then you put this box into a tent, and you take  
5 very, very low food reserves or if it's good weather, you can  
6 even do no food reserves, wait one or two days and then you put  
7 the queen frame in. Before this or one day before you put the  
8 queen frame in, you make your application so you ensure that the  
9 bees have collected nectar and pollen already.

10                   So, you know that that pollen and nectar  
11 inside is already contaminated and that they will be forced  
12 during the rearing of the queen to forage on this contaminated  
13 crop. Then, that is actually quite a short process until the  
14 queen cell is capped. You will definitely have the flowering  
15 period still, and then you can take them out and you can observe  
16 them, because then the queen is not fed anymore and then you can  
17 do additional experiments to the queen if you would like, but  
18 that's actually quite short and quite efficient test I would  
19 say. I don't know.

20                   **DR. DANIEL SCHLENK:** Dr. Hunt?

21                   **DR. GREG HUNT:** I agree.

22                   **DR. DANIEL SCHLENK:** Dr. Pettis?

23                   **DR. JEFF PETTIS:** Jeff Pettis. And I all  
24 ready want to modify the method. No. You don't like feeding  
25 nectar and spiked pollen, but I think this might be a perfect -  
26 rather than have them forage on the crop, if you know something



1 about the residue in the nectar and pollen, why not just  
2 simulate that in a shook swarm, and just keep them completely  
3 artificial in little cages by themselves and introduce the  
4 queen. I agree, it's a great idea that asking a group of  
5 queenless bees to rear queen cells should be a great bioassay.

6 **DR. DANIEL SCHLENK:** Mr. Pistorius?

7 **MR. JENS PISTORIUS:** And I agree with Dr.  
8 Pettis. In this case, I would actually think if you know the  
9 concentrations that you have to spike nectar and pollen, that's  
10 a great idea then in a confined tunnel condition.

11 **DR. DANIEL SCHLENK:** Dr. James?

12 **DR. ROSALIND JAMES:** I don't know if this  
13 would really be considered a Tier II test. Throughout this,  
14 I've been wondering could you require at the Tier II level that  
15 a field experiment be conducted without bees in which then you  
16 measure actual residues in the pollen and nectar from different  
17 types of applications. That will give you some of these unknown  
18 answers, well how much then do we feed to bee.

19 **DR. DANIEL SCHLENK:** Okay. Anyone else?  
20 Okay. Let me go to the agency. Any questions of clarification?  
21 We're good? Okay. Let's move onto letter C.

22 **MR. JOSEPH DECANT:** Joe DeCant, question 11,  
23 subpart (c) - please comment on the most important endpoints  
24 that should be measured in the Tier II studies, such as adult  
25 forage bee mortality, brood development, queen fecundity,  
26 overall colony strength, that are linked to assessment endpoints



1 and their associated protection goals.

2 **DR. DANIEL SCHLENK:** Dr. Tarpy?

3 **DR. DAVID TARPY:** This is Dave Tarpy. We  
4 discussed this briefly and largely agreed that those that were  
5 outlined in the White Paper were very adequate at looking at  
6 amount of brood, the overall colony population and strength, all  
7 of those measures of colony phenotype are very appropriate, and  
8 are a good way to quantify colony level effects. I should note

9 I should note that by and large, all of those  
10 different measurements are highly correlated in and of  
11 themselves. So, quantifying a composite colony phenotype in  
12 these ways can be done statistically, principle component  
13 analyses, using different loading variables into that is a way  
14 to - is a means of comparison and is quite effective in doing  
15 that.

16 To touch on the points that were just raised  
17 about looking at queen and don't forget drones - right, drones  
18 as well as queens - queen and drone viability and their  
19 individual phenotypes is another outcome and endpoint that  
20 should be measured in these types of Tier II studies as well.

21 A third and final major endpoint is to sample  
22 and process returning foragers and to actually see what the  
23 foraging behavior is, quantifying their exposure rates,  
24 quantifying what their foraging on, and looking at the returning  
25 foraging force in these Tier II studies to really quantify their  
26 potential exposure routes.



1                   **DR. DANIEL SCHLENK:** Okay. Dr. Ostiguy?

2                   **DR. NANCY OSTIGUY:** I think this is also a  
3 point where some in-hive samples could be taken to potentially  
4 supplement. We had information on the contamination at the  
5 pollen and nectar to then look at the corresponding  
6 concentrations of the pesticides within honey and bee bread.

7                   **DR. DANIEL SCHLENK:** And Dr. Pettis?

8                   **DR. JEFF PETTIS:** To extend what Nancy said  
9 just a bit further, at the end of the exposure period within a  
10 tunnel in addition to looking at bee bread an pollen, you can  
11 take newly emerged adult bees that have been fed and processed  
12 during that exposure period and assay them directly, or then put  
13 them in cages and do longevity studies. A fairly easy thing to  
14 do, at the end of the study, doesn't add much to the cost.

15                  **DR. DANIEL SCHLENK:** Dr. James?

16                  **DR. ROSALIND JAMES:** Question for Dr. Tarpy.  
17 So, you said sample then process returning foragers to see what  
18 they're foraging on, but in a tunnel study, we pretty much know  
19 what they're foraging on, no?

20                  **DR. DAVID TARPY:** Sorry, this is Dave Tarpy.  
21 One can quantify the nectar loads of individual bees by  
22 capturing them at entrances, anesthetizing them with carbon  
23 dioxide, then squeezing their crop contents into capillary  
24 tubes, then very accurately measuring the nectar loads of each  
25 forager and then sampling many foragers to get an idea of the  
26 variation in how many are foraging on nectar versus pollen, what



1 the average nectar loads are. You can even look at sugar  
2 content and other things. This is again, very well documented  
3 in the literature. That's what I meant is actually to sample  
4 the incoming foragers to see kind of more quantitatively of what  
5 their exposure rates are.

6 **DR. DANIEL SCHLENK:** Okay. Any other Panel  
7 members? Mr. Pistorius?

8 **MR. JENS PISTORIUS:** Real short - for me, this  
9 list is missing behavior at the hive entrance and behavior of  
10 foraging bees on the crop.

11 **DR. DANIEL SCHLENK:** Okay. Make sure that we  
12 get that on the list. Okay. It seems pretty straight forward,  
13 but I'll ask if you guys have any questions of clarification?  
14 Yeah, Mr. Sappington?

15 **MR. KEITH SAPPINGTON:** Keith Sappington.  
16 Quick question regarding the nectar loads in measuring those.  
17 Would that include measurement of residues in the nectar as well  
18 from the tunnel studies?

19 **DR. DAVID TARPY:** This is David Tarpy. I  
20 assume that that could be done as part of that, yeah.

21 **DR. DANIEL SCHLENK:** Okay. Let's go ahead and  
22 move onto letter D.

23 **MR. JOSEPH DECANT:** Joe DeCant, question 11,  
24 subpart (d) - section 4.2.2 of the White Paper discusses a  
25 full-field feeding design. This methodology is discussed under  
26 Tier II assessments since the colony is relatively confined to



1 foraging on either spiked sucrose solutions or spikes pollen.  
2 The intent of this methodology is to ensure that colonies are  
3 exposed to known residue levels over longer durations than the  
4 semi-field tunnel study designs. A limitation to the study is  
5 that bees may simply store spiked food rather than consume it  
6 and that the reliance on a single source of food may introduce  
7 confounding effects, such as nutritional deficits into the  
8 study. Please comment on the environmental realism and utility  
9 of full-field feeding studies as a line of evidence in  
10 characterizing risk to honey bee colonies.

11 **DR. DANIEL SCHLENK:** Okay. Dr. Tarpy?

12 **DR. DAVID TARPY:** So, I feel like we've all  
13 ready kind of discussed this quite a bit. So, I don't know if  
14 my comments here are really going to be all the relevant, but I  
15 guess there is some disagreement on the panel here as to whether  
16 is kind of zero utility, some utility or a lot of utility. I  
17 guess what it comes down to is that a well-designed full-field  
18 Tier II study, I think, would provide information that would not  
19 be gleaned at a Tier I level. Again, it may not realistically  
20 mimic a full ecological type of realism, but I believe it would  
21 have some - this type of design would have some utility.

22 Again, honey bee colonies, I would think,  
23 compared to tunnel studies where there are some limitations just  
24 on their behavior being amenable to being in small enclosures  
25 that they would be more amenable to these full-field types of  
26 designs. I think that by being in enclosures, one of the



1 limitations that we discussed earlier in this question is the  
2 shortened duration where as a full-field design would facilitate  
3 longer measurement durations of colonies and for chronic levels  
4 of exposure that could be very beneficial, especially looking at  
5 colony population and turnover where brood raised under a  
6 particular exposure could then be measured as adults.

7 So, extending it beyond 28 days, which I  
8 believe the White Paper was mentioning, spending it beyond that  
9 to try and quantify some of those phenotypes of individual bees  
10 that were raised under those particular environmental  
11 conditions.

12 **DR. DANIEL SCHLENK:** Okay. Dr. Ostiguy?

13 **DR. NANCY OSTIGUY:** One possible way to  
14 address the issue about whether or not the bees store the  
15 contaminated material and not use it to raise current brood  
16 would be to actually sample the colony to require some  
17 indication of what was in the colony in the first place in terms  
18 of brood food, honey, et cetera, and then have a measurement  
19 that told you how much of what was used would possibly have been  
20 from the contaminated material.

21 **DR. DANIEL SCHLENK:** And, Dr. Pettis?

22 **DR. JEFF PETTIS:** Recognizing the concerns of  
23 my esteemed colleague, Mr. Pistorius, I actually do like these  
24 studies. If we recognize it, they may not represent the contact  
25 element. I think given that we reduce the stores at the  
26 beginning of the study and then spike them with relative rates



1 in nectar and pollen that they do have some value. Again,  
2 running them at a time of year when there is less incoming  
3 resources, so you have some control over the consumption rate.  
4 We have a distinct spring and fall pollen and nectar flow. So,  
5 there are times in the year when they can be done.

6 **DR. DANIEL SCHLENK:** Dr. Berenbaum?

7 **DR. MAY BERENBAUM:** I think just as integral  
8 to the inclination of bees to forage broadly geographically is  
9 their tendency to glean. One missing worry here - there is a  
10 concern about nutritional deficits and storage of compounds, is  
11 that force feeding eliminates that behavior. It's an incredibly  
12 sophisticated system of communication, decision making, force  
13 feeding does not capture that flexibility in bee behavior. So,  
14 yes, these force feeding studies can be informative if and only  
15 if there is an understanding of the natural disposition of bees  
16 to learn a verse of stimuli and avoid them in the future and to  
17 sample and detect and possibly chose not to feed. My experience  
18 is mostly with caterpillars and even caterpillars, when given a  
19 choice, can avoid toxic food. It's much more difficult to  
20 interpret a no choice test than a choice test in terms of its  
21 ecological reality.

22 **DR. DANIEL SCHLENK:** Thanks. Dr. Pettis?

23 **DR. JEFF PETTIS:** Good point, May. I guess in  
24 some of the studies that I've observed or been a part of, you  
25 look at consumption rate by the colonies and often they are  
26 equal, so you can at least - you know, again, they're not given



1 a choice. You have a spiked syrup and non-spiked. At least  
2 there is no deterrent to feeding or obvious deterrent.

3 **DR. DANIEL SCHLENK:** Mr. Pistorius, you have  
4 something to add on this one?

5 **MR. JENS PISTORIUS:** I will add a little bit.  
6 Just from feeding studies, I think we have less of, for  
7 instance, repellency effect with feeding sugar solutions. So,  
8 you can actually - and maybe this is, you know, for some  
9 specific questions, but it should not be the only one for your  
10 lines of evidence if you have a good OECD and EPPO study. You  
11 can have such studies as additional information, but for  
12 instance, we know from all the laboratory toxicity tests that  
13 the bees will feed on this. Even with pyrethroids, which have  
14 repellency effect when you spray them on flowering crops, if you  
15 give a sugar solution to the colony, they will just consume it.  
16 They'll take whatever is there.

17 **DR. DANIEL SCHLENK:** I hear Mary Poppins in  
18 there somewhere right? Any other comments on section D here?  
19 Yes, Dr. Hunt?

20 **DR. GREG HUNT:** Well, I think that when you do  
21 the experiment this way in the field and feeding, you don't know  
22 what the dose is. You don't really know what the dose is, but  
23 possibly it could be useful for, as Dr. Tarpy said, for looking  
24 at that cohort of bees that was raised during the time you're  
25 feeding and they're receiving some unknown dose, field relevant  
26 dose, and see if there're effects.



1                   **DR. DANIEL SCHLENK:** Okay. Any other comments  
2 on letter D. Okay. Let me go to the Agency. Mr. DeCant, do  
3 you have any questions of clarification?

4                   **MR. JOSEPH DECANT:** Joe DeCant, EFED. I have  
5 one question of clarification. If we're talking about this  
6 field feeding design and we're talking about the cohort that was  
7 raised under those conditions and we talked about the ability of  
8 the bees to store pollen and nectar, to what extent does the  
9 Panel agree about just that cohort of bees or the actual  
10 duration of the study? I guess there's a question of the  
11 duration of the study when bees can store the food and consume  
12 it later.

13                  **DR. DANIEL SCHLENK:** Dr. Tarpy, you want to  
14 lead off on that?

15                  **DR. DAVID TARPY:** Is the question about what  
16 the duration should be? Or which cohort you're asking?

17                  **MR. JOSEPH DECANT:** Joe DeCant. I think it  
18 would be about which cohort to look at, which also relates to  
19 the duration of the study. And I say that because, you know,  
20 we're talking about measurement endpoints and I think - so the  
21 White Paper discusses about minimizing the number of times that  
22 we have to open up the hive to collect our measurement  
23 endpoints. So, I think if the Panel has any insight about what  
24 cohort to target, maybe a timing of the study if we're talking  
25 about the dearth period, if that's in the middle of the summer  
26 time and there is a honey flow in the spring and the fall, you



1 know the duration of the study - and I guess ultimately this  
2 gets back to maybe a clarification about under what conditions  
3 would this type of study be most useful. If we're talking about  
4 longer duration, the measurement endpoints, when to collect  
5 those measurement endpoints and the cohort to target as you  
6 mentioned about, Dr. Tarpy.

7 **DR. DAVID TARPY:** Okay. This is David Tarpy.  
8 Others feel free to chime in here, but I think that there are  
9 many cohorts that would be informative to this type of design.  
10 One would be those active foragers and nurse bees. So, say  
11 30-day-old bees and 10-day-old bees, during the time of  
12 exposure, during the time of feeding, and then several weeks  
13 later, emerging, marking and following of the cohort of bees  
14 that were larvae during the time of that exposure as well.

15 So, minimizing the number of times going into  
16 the colony, I don't see that as a limitation or a problem. I  
17 just see that all of the different cohorts would be very  
18 informative in their different ways. So, spending the period of  
19 time - and plus you could go on for a very long period of time  
20 looking at then subsequent cohorts as well.

21 So, there really is no definitive endpoint to  
22 this, it just comes down to tracking these things over time. I  
23 don't know if there is one particular time period that makes  
24 more sense than others. Certainly, just looking at one period  
25 of time is going to be limited in scope.

26 **DR. DANIEL SCHLENK:** Dr. Pettis?



1                   **DR. JEFF PETTIS:** Kind of echo what David  
2 said. If you have a 20-day flowering period and you have  
3 colonies even in a tunnel study, you want to look at adults  
4 right during the flowering period, their behavior and affect on  
5 adult mortality. But this brood you want to target maybe the  
6 midpoint of the flowering period, 10 days in, as a start point.  
7 You need those nurse bees to process that food for about 10 days  
8 before they actually, before their glands are fully developed  
9 and they're feeding larvae.

10                   So, then you just keep moving the clock.  
11 We've been going out about 30 days beyond the midpoint of the  
12 flowering period to capture that. And of course, if you have  
13 opportunity to follow multiple cohorts, it's better. You may  
14 see during the peak of incoming nectar and pollen, you may see  
15 an effect. Then later, you may see that effect disappear. So,  
16 following multiple cohorts - I noticed in the White Paper that  
17 there was a concern about disruption to the colony. I think  
18 given very good beekeeping practices, that's not such an issue.  
19 You will see sometimes a spike in adult mortality if the person  
20 managing the hives is not careful, but I think the need to get  
21 the data probably overweighs and just hiring a good beekeeper is  
22 a way to avoid that.

23                   **DR. DANIEL SCHLENK:** Anybody else want to  
24 chime in on this one? Dr. Hunt?

25                   **DR. GREG HUNT:** Well, I think that some of  
26 this information can be gotten out of tunnel studies. I think



1 there is some kind of consensus that tunnel studies are good,  
2 more controlled and should be the main focus of Tier II. If  
3 anybody disagrees, speak up now.

4 **DR. DANIEL SCHLENK:** Yeah, in terms of writing  
5 this up, it sounds like it would be nice to present just exactly  
6 what you said. It seems like some people are for them, limited  
7 and the caveats as well as those, the reasons why they're not  
8 useful, I guess that would probably be a good way to put that in  
9 the minutes. Would that be helpful? Yeah? Dr. Pettis?

10 **DR. JEFF PETTIS:** I guess along that line, do  
11 we feel as a group that the free-flying feeding studies are not  
12 - like you feel that they're not Tier II at all, they're really  
13 Tier III studies. Is there is any general consensus among the  
14 group about whether free-flying colony level studies with spiked  
15 stuff, is that more of a Tier III or more Tier II? Is there a  
16 feeling?

17 **DR. DANIEL SCHLENK:** How many say it's a Tier  
18 III? Okay how many IIs? Look like we're split. So, sounds  
19 like it's a split. Looks pretty even actually. Pardon? Yes,  
20 you can clarify, Mr. Pistorius.

21 **MR. JENS PISTORIUS:** With respect to what is  
22 the ultimate possibility for decision? So I guess it would  
23 actually be rather a Tier II. That was not my point. I think  
24 the study types are not the only ones that you can rely on and  
25 it was basically the study design that I was criticizing, but  
26 now in the discussions, I think we have elaborated some very few



possibilities where actually a feeding study can also be useful or in addition to a really good OECD, EPPO, you can use this feeding to enhance. I hope that clarifies.

**DR. DANIEL SCHLENK:** I'm sure we'll look forward to the written comments on this one. Dr. Hunt?

**DR. GREG HUNT:** I want to change my vote. Yes, it's more of a Tier II, and clearly the experiment with raising queens would be Tier II because they're not free flying.

**DR. DANIEL SCHLENK:** Okay. Noted. I guess that I think there is - are we II as well?

**DR. JEFF PETTIS:** Last comment - I've all ready cast my vote. Last comment along these lines, I do feel that the EPPO guidelines and the OECD - there was a lot of thought and effort that goes into them. They are very valid things to follow.

**DR. DANIEL SCHLENK:** Yeah, we can set this up in the minutes in terms of kind of a split decision and people can weigh in before it actually comes out at the end to get a final vote, whatever that is, whatever that means for you guys. Okay. Any other comments on this? Mr. DeCant, are you fine with what we've given you so far on that at this point? Okay. You want to read in letter E then?

**MR. JOSEPH DECANT:** Joe DeCant, question 11, subpart (e) - as discussed in section 4.3.4 of the White Paper, it is important to consider the biological significance of a measured effect in addition to its statistical significance.



1 Please comment on the nature and magnitude of effects that would  
2 be sufficient to conclude biologically significant effects on  
3 the colony and/or the need to transition to Tier III  
4 assessments.

5 **DR. DANIEL SCHLENK:** Dr. Tarpy?

6 **DR. DAVID TARPY:** This is David Tarpy. I  
7 think we agree 100 percent that reconciling statistical and  
8 biological significance can be a challenge. I would probably  
9 just want to leave it at that. This is an incredibly difficult  
10 question to answer with any sort of certainty. I mean, this is  
11 something that we all wrestle with of course, and it's even  
12 further blurred and confused by this kind of social nature of  
13 honey bees, and it just makes it even more difficult of a  
14 challenge because what can be very, very small, seemingly  
15 insignificant effects at the individual level can have profound  
16 ramifications at the colony level.

17 Conversely, very large, seemingly striking  
18 effects between individuals can be balanced out and can be  
19 compensated for at the colony level. So, it is really, really  
20 difficult to try to reconcile this. So, I don't know if we're  
21 going to really be able to do justice to this particular  
22 question, but all of that said, we believe that power analyses  
23 that was asked for in the White Paper in multiple occasions at  
24 these different tiers feel like that's incredibly informative  
25 and should definitely be included in all of the results.

26 This is especially true with the small sample



1 sizes, small colony sample sizes because of the inherent  
2 variability across colonies in different contexts, different  
3 environments with larger variability, higher sample size is  
4 needed really to be able to elucidate statistically significant  
5 effects, which hopefully will provide inference to biologically  
6 meaningful effects. So, larger sample size is obviously a  
7 logistical limitation, but it's one that needs to be maximized  
8 with a sense of urgency, you know, at all possibility.

9           There were some comments earlier talking about  
10 the Tier II studies and different measurements that would be  
11 taken. One that touches on this particular question about kind  
12 of teasing apart statistical versus biological relevant effects  
13 is doing ground truthing and looking at actual foraging behavior  
14 in the target crop or whatever the bees are actually foraging  
15 on. Those type of data, I think, would really help augment the  
16 interpretation of trying to tease apart statistical versus  
17 biological effects.

18           Some other points here that I think we've  
19 already raised, so I won't go over those again. I think one  
20 last approach is trying to merge data sets or trying to  
21 consolidate data. One difficulty of having low sample sizes and  
22 lower power, if different data sets could be combined and have a  
23 meta analysis approach, that is a way to also reconcile this  
24 point that there may be small, even statistically nonsignificant  
25 effects that aren't detected within a single study, but by  
26 bolstering sample size by doing this similar study in different



1 environments and different contexts, meta analyses can often  
2 pull out biologically relevant effects where each individual  
3 study might not have statistically meaningful effects.

4 So, that again might be beyond the scope of  
5 this, but that is another avenue to consider in consolidating  
6 data.

7 **DR. DANIEL SCHLENK:** Thanks. Dr. Ostiguy?

8 **DR. NANCY OSTIGUY:** I'm going to make another  
9 plug for the analytical models because you can get two things  
10 from them. One data that may not show statistical significance  
11 or variables that may not show statistical significance may turn  
12 out to be incredibly biologically important. You may have  
13 somethings that the analytical model would tell you that you  
14 have statistical significance on that variable, but it doesn't  
15 really matter for the colony.

16 **DR. DANIEL SCHLENK:** And Dr. Pettis?

17 **DR. JEFF PETTIS:** Just very briefly, again we  
18 all feel better when we have high statistical power and we have  
19 good P values that lead to better publications and stuff like  
20 that. But, I don't think we can discount some of the biological  
21 effects that we see and I'll give you an example.

22 So, you have a Tier I study, you're measuring  
23 mortality and other things, but you notice that the bees are  
24 excessively grooming, which could mean a response to pesticides.  
25 Then you notice the same response in a Tier II or a Tier III  
26 study. So, you see an effect, but it's statistically not valid.



1 I think these biological effects that we see, we have to take  
2 them into consideration and try to figure out what they mean for  
3 the colony level. But we recognize that for many of these  
4 colony level studies, the ability to do adequate replication is  
5 hard to get there - economically it's hard to design a study  
6 well enough and have a enough replication to strong statistical  
7 power.

8 **DR. DANIEL SCHLENK:** Okay. Any other panel  
9 members want to weigh in on this one? Dr. Hunt?

10 **DR. GREG HUNT:** Just a comment. You're  
11 dealing with a bunch of academics here. So modeling and meta  
12 analyses are maybe good for discovery and for planning, but to  
13 echo what Jeff said, you know, I guess what we're really talking  
14 about is testing for risk analysis. So, there're two different  
15 things going on here.

16 **DR. DANIEL SCHLENK:** Dr. Fefferman?

17 **DR. NINA FEFFERMAN:** While modeling - so, I'm  
18 not going to speak so much to the meta analysis for statistics  
19 because honestly, I'm not a biostatistician, but while modeling  
20 has a huge role in discovery, it can also lead to  
21 recommendations for which things to test as measurements, and  
22 what those thresholds for the tests should be. So, I think it  
23 would be a mistake to dismiss as a purely academic concern that  
24 doesn't directly lead to applied recommendations.

25 **Dr. DANIEL SCHLENK:** Any other comments?  
26 Okay. Go to the Agency, any questions of clarification? Keith?



1  
2 **MR. KEITH SAPPINGTON:** Keith Sappington.

3 Yeah, this is a difficult question and it again, cuts across  
4 taxa, the biological and statistical significance and it would  
5 obviously differ depending on endpoints.

6 Speaking from my own experience in reviewing  
7 half a dozen or so semi-field studies, I understand there is  
8 going to be a lot of gray area where best professional judgement  
9 is needed, but if there are bounds if we could put on, if  
10 there're areas where you say, look this is an obvious place  
11 where its biologically significant because we're dealing with  
12 endpoints like brood compensation index and brood termination  
13 rates as well as percent comb areas and various stages of brood.

14 So to help us with that interpretation, I'm just speaking to  
15 the point of biological significance, that would be extremely  
16 helpful.

17 **DR. DANIEL SCHLENK:** So if I may, it sounds  
18 like which biological endpoints have a more biological relevance  
19 than say a statistical relevance out of the ones that you're  
20 sort of measuring. Is that.

21 **MR. KEITH SAPPINGTON:** Keith Sappington.

22 Partly, yes, which endpoints maybe carry a little more  
23 biological weight, but even within an endpoint. For example,  
24 with the OECD studies 75, there are endpoints including brood  
25 termination rate and so forth, but there isn't, to my knowledge,  
26 a criterion of what's acceptable for that in the controls. So,



1 there's interpretation around that. And then just understanding  
2 the biological significance, so if you do have a two-fold change  
3 regardless of what the stats can say, is that meaningful? And  
4 again, obviously that can get into the modeling, which we will  
5 be talking about, but any insights based on your experience with  
6 conducting these studies and knowledge of bee biology would be  
7 helpful. Thank you.

8 **DR. DANIEL SCHLENK:** Dr. Tarpy, you want to  
9 come up with some guesses there?

10 **DR. DAVID TARPY:** So would it be helpful to  
11 the Agency - sorry this is Dave Tarpy again - would it be  
12 helpful to perhaps rank or somehow qualify the list of different  
13 colony phenotypes on page 132 and 133 in the White Paper to say  
14 as biologically critical versus more environmentally  
15 temperamental or something like that where that would give you  
16 an idea of the gravity of the particular measurement, is that  
17 kind of what you're looking at as far as which are more  
18 biologically relevant versus less?

19 **MR. KEITH SAPPINGTON:** Keith Sappington. Yes,  
20 if that can be done with some confidence, then that would be  
21 helpful, sort of a first tier versus maybe a second tier in  
22 terms of biological significance. But again, even within an  
23 endpoint, understanding daily mortality rates while I have a  
24 six-fold increase for three days, but then things return back to  
25 normal. I'm not asking for a specific answer on just that, but  
26 understanding even within an endpoint if there are bounds and



1 very clear bounds on the magnitude that says yes, this is a  
2 problem. Like we had discussed earlier that perhaps a sustained  
3 mortality of 30 percent based on experience, it looks pretty  
4 major. So, if there are some bounds that we can put around the  
5 zone of uncertainty, that would be helpful, thank you.

6 **DR. DANIEL SCHLENK:** Okay. We'll go with Dr.  
7 Berenbaum first and then Dr. Fefferman.

8 **DR. MAY BERENBAUM:** Completely shooting from  
9 the hip, in as much as I have never done any studies. But just  
10 in terms of statistics and biological relevance, it would seem  
11 those parameters that relate to the queen who is not replicated  
12 in any colony should be given some latitude in terms of  
13 biological reality. Also, given the incredibly central role of  
14 the queen, you have on page 132 there are some measurements  
15 relating the presence of the same queen. I mean, within a  
16 single colony, you don't have that same statistical power that  
17 you do with even a small colony with 5 or 10,000 workers. So, I  
18 would say you should have some sort of - if something goes wrong  
19 with the queen and that is biologically relevant, let the bee  
20 biologists address that one.

21 **DR. DANIEL SCHLENK:** Dr. Fefferman?

22 **DR. NINA FEFFERMAN:** So I think we're  
23 conflating two different aspects into the same conversation, and  
24 one is - we're conflating two different elements. One is how to  
25 get from the statistical significance to the biological  
26 significance and the other is the criticality of the biological



1 significance.

2 I think Dr. Berenbaum's statements speak  
3 directly to how to get from the statistical to the biological,  
4 but it would be a mistake to try and intuit which aspects are  
5 critical biologically relevant aspects as opposed to transient  
6 biologically relevant aspects. Within the category of being  
7 biologically relevant just isn't off the cuff because we've got  
8 example after example from the last 50 to 100 years of research,  
9 of experts in the field with very good reasons and very sound  
10 science behind them going, I really think this is the important  
11 thing, later testing that in either modeling or as greater  
12 sensitivity of empirical techniques become available, realizing  
13 that the intuition is in fact really off.

14 So, while we can do that, I think we have to  
15 separate the discussion from the statistical to the biologically  
16 relevant and then be very careful within the biological  
17 relevance of estimating relative criticality.

18 **DR. DANIEL SCHLENK:** Okay. Dr. Ostiguy?

19 **DR. NANCY OSTIGUY:** I'd like to add that in  
20 addition to queens, if something goes wrong with your drone  
21 population, you're in trouble.

22 **DR. DANIEL SCHLENK:** Anybody want to add the  
23 other ones too? Dr. Pettis?

24 **DR. JEFF PETTIS:** I'm glad to hear that female  
25 side speaking up for drones. That's encouraging. Not Greg, are  
26 we looking at here.



1                   **DR. DANIEL SCHLENK:** Except for Greg here.

2 Greg's kind of.

3                   **DR. JEFF PETTIS:** Back to biological  
4 significance, even with the comment of Dr. Fefferman, I think  
5 one thing that I look at and Dave, I think we could take some  
6 stab at trying to prioritize to some degree, maybe in two  
7 categories. The one that jumps out at me always is just the  
8 level of brood production and brood survival. The reason that  
9 is so important is it incorporates so many other aspects of the  
10 hive activity. Incoming pollen and nectar, nurse bee activity,  
11 it really is a great measure.

12                   When we look - if we're measuring colony  
13 strength, we can measure adult population, which is harder to  
14 measure, but we can measure sealed brood production. It's real  
15 easy to measure very accurately as a very robust measure. So,  
16 if I had to look to one measure, it would be something about  
17 brood survival or brood production. But again, I agree that we  
18 can't over look everything that is out there. We have to  
19 consider that it is perhaps important.

20                   **DR. DANIEL SCHLENK:** Okay. Anybody else got  
21 another favorite measure? Yes, Mr. Pistorius?

22                   **MR. JENS PISTORIUS:** I was keeping back,  
23 because it is such a hard question and we ask ourselves a lot  
24 too. It depends on the level of protection that you want to  
25 achieve for your protection goals. If your major protection  
26 goal is the colony size, okay then you might have forager



1 mortality event, which is not that relevant for colony survival  
2 and colony development over the long term.

3 On the other hand, we are for instance,  
4 incident driven. We don't want incidents because that is part  
5 of my job, investigating the incidents. Don't need too much  
6 work. What we don't want, for instance, acute mortality events.

7 So for us, it is a concern if we have clearly increased - it is  
8 again the question what level. But if we clearly increase  
9 mortality, there was a saying from a bird person from the  
10 Netherlands who said in the Netherlands, they don't want clearly  
11 visible dead birds. We want exactly the same for bees. We  
12 don't want clearly visible dead bees. That's why with the  
13 methodology of the bee trap we are a lot more protective  
14 compared to what the beekeeper actually realizes.

15 If we see a clear increase in mortality, we  
16 even say okay, this is for us biologically relevant, even if the  
17 brood production is not yet affected. So for me personally, the  
18 most critical, in my personal top list, is acute mortality.  
19 Then I agree, it is not really below it on the same very  
20 important level is the brood production. Because depending on  
21 the substance, if you have an IGR, of course you would not get a  
22 clearly visible incident, but you will have no more brood. So,  
23 they are both basically on the same level. They are the most  
24 important ones in my opinion.

25 Foraging behavior, okay this is also very  
26 important. For instance, if you would have a substance where



1 you have a clear impact on and a long lasting impact on the wish  
2 of the bee to forage on that crop, that would of course affect  
3 one of your protection goals too, the quantity of hive products.

4 So, I think this is also very important. We have to take them  
5 all into account. Ranking will be difficult, but those are I  
6 think the most relevant ones.

7 **DR. DANIEL SCHLENK:** Dr. Berenbaum?

8 **DR. MAY BERENBAUM:** I think maybe splitting  
9 hairs to say what's most or least relevant. But there's one  
10 profound difference in assessing nontarget impacts on bees than  
11 for birds or most other nontarget arthropods is that most  
12 people, even though the nontarget organisms can be beneficial.  
13 Most people don't make a living out of those beneficial  
14 organisms.

15 So, in the case of nontarget impacts on bees,  
16 it's important economically not only that they are not dead, but  
17 that they are also functional and capable of both delivering the  
18 economic services for which they are being raised. So, it's a  
19 different context. I used the analogy earlier in a conversation  
20 in some ways and army, a dead soldier - no disrespect to the  
21 military - but a dead soldier at least is easier to deal with  
22 than one that's suffering mental or physical problems.

23 So, in a way, we have these troops out there  
24 that are delivering pollination services. So, it's not just  
25 that their numbers are reduced. If their behaviors are  
26 profoundly altered so that they are not delivering the



1 ecological and economic services, for which they are being  
2 maintained. That's important in the context of risk assessment.

3  
4 **DR. DANIEL SCHLENK:** Dr. James?

5 **DR. ROSALIND JAMES:** A point taken with the  
6 answer, you could go back to your protection goal and say what  
7 are your three protection goals and weigh them. So, what are  
8 they provision of pollination services, so what is going to be  
9 important for provision of pollination service. I will be hive  
10 activity but also be colony size and colony survival. That goes  
11 back to what you were saying about protecting the beekeeper  
12 also, and then similar for production of hive products will be  
13 the same factors, generally colony size and colony survival.

14 **DR. DANIEL SCHLENK:** Dr. Pettis?

15 **DR. JEFF PETTIS:** You beat me to it, Rosalind.  
16 But going back to those two protection goals, the first two  
17 protection goals, your point about loss of forager force in a  
18 pollination contract could be critical. So, a short impact on  
19 pollination or the pollinating force would be critical at that  
20 point, even though it may not have colony level effects, which  
21 would affect whatever protection goal number two.

22 **DR. DANIEL SCHLENK:** Dr. James?

23 **DR. ROSALIND JAMES:** But then I still have to  
24 add - the fecundity still keeps coming back. We had a case with  
25 a growth regulator and alfalfa leaf cutting bees. So a new  
26 growth regulator was approved for alfalfa, applications to



1 alfalfa seed and after it was used in fields the first year we  
2 had the beekeepers as alfalfa leaf cutting beekeepers come to us  
3 and say we got very poor bee return, that is the number of bees  
4 that came back out of the field, from those fields that were  
5 treated with this product and be return was normal in the fields  
6 where the product wasn't used. The product had very low acute  
7 toxicity to adult bees.

8 So then, some of the scientists in our group  
9 investigated it and well, if it's a growth regulator, it's not  
10 going to really affect adults, but maybe it'll affect fecundity.

11 It turned out a closer investigation that the queens were  
12 laying inviable eggs, either that or the larvae exposed to the  
13 pollen provision was contaminated. We don't know which, but the  
14 eggs were not hatching or when they hatched, they died  
15 immediately. So, fecundity kind of comes into play always also.

16  
17 **DR. DANIEL SCHLENK:** Dr. Fefferman?

18 **DR. NINA FEFFERMAN:** So, in some sense, I love  
19 all of these comments and I'm now almost stealing my own  
20 thunder, because you asked me to put examples into question 14.  
21 This was the example that I put in. We've been talking about  
22 rates of mortality as an issue and now we're talking about which  
23 things are most critical. It's a mistake to separate those two.  
24 Mathematically, if you had an analytic model for this, it is  
25 the ratio of worker death to worker recruitment. Whether that's  
26 from eclosion or egg laying, whatever is driving that, that



1 ratio has to be greater than one in order for the colony to be  
2 successfully stable as a population demographic. And either  
3 side of that, either because the eclosion rates drop or because  
4 mortality goes up can drive that ratio less than one.

5 That is why I'm really sort of cringing about  
6 identify these set of things as being critical because neither  
7 of those in isolation is necessarily critical to colony  
8 survival. It is critical - I like very much Dr. Pettis's idea  
9 if it is critical to the pollination services over a duration,  
10 but in terms of colony survival, neither one in isolation is  
11 itself critical. The ratio of the two is incredibly critical,  
12 and that is the sort of thing where just shooting from the hip  
13 on what seems to be critical gives you sort of the wrong insight  
14 into that.

15 **DR. DANIEL SCHLENK:** So, let me just remind  
16 the Panel again that anything that you say, you need to get  
17 written comments to Dr. Tarpy on this particular issue, because  
18 he needs to put these together into some coherency. So, just a  
19 reminder there. Any other comments? Dr. Hunt?

20 **DR. GREG HUNT:** Maybe I'll get the last word.  
21 We can't really rank these things as has been said, but anything  
22 the affects the reproductives would probably be at the top.  
23 Like if you get deformed queens, that's bad.

24 **DR. DANIEL SCHLENK:** Okay. I think you're  
25 going to get a list, Keith, on a bunch of stuff.

26 **MR. KEITH SAPPINGTON:** Thank you very much.



1                   **DR. DANIEL SCHLENK:** Any other questions,  
2 clarification questions from the Agency? Alright, let's take a  
3 break. Let's come back at a quarter to three.

4                   (WHEREUPON a recess was taken)

5                   We're ready to lead off with question 12,  
6 letter A.

7                   **MR. JOSEPH DECANT:** Joe DeCant, EFED, question  
8 12, subpart (a) - please comment on the strengths and  
9 limitations of full-field studies described in the white paper.

10                  **DR. DANIEL SCHLENK:** All right, Dr. Pettis,  
11 you're our lead discussant.

12                  **DR. JEFF PETTIS:** Thank you -- Dr. Pettis.  
13 Tier III studies are meant to be ultimate tests of real world  
14 exposure and potential effects. However, these studies are  
15 fraught with potential pitfalls with regard to the ability to  
16 carry out controlled experiments in the open environment and  
17 with adequate controls.

18                  When properly executed, Tier III studies are  
19 the best means to assess the risk and impacts of pesticides to  
20 Apis mellifera colonies and other managed colonies such as  
21 Bombus or Osmia. In the context of the regulatory framework,  
22 Tier III studies are considered highly refined from the White  
23 Paper. Studies meant to address specific concerns raised in  
24 lower tier studies, in that said, each study design should vary  
25 with regard to the endpoints that we measured to address these  
26 specific concerns that may be raised in Tier I or Tier II



1 testing.

2 So, I will point out some strengths and some  
3 weaknesses that the group felt were relative. So for strengths,  
4 the natural exposure that can be achieved by allowing bees to  
5 forage freely on the treated crop and then measuring endpoints  
6 allows for the testing of effects at the colony level that are  
7 unattainable at Tier I and Tier II level testing. The exposure  
8 achieved can accurately simulate the actual use of the  
9 pesticide, another strength.

10 Tier III tests allow for colony-level testing  
11 of protection goals. So, it's the only aspect. Maybe a little  
12 bit in Tier II, but you can do colony-level test. If  
13 significant effects are noted at the colony level, the results  
14 can be interpreted with a high level of confidence as the colony  
15 has many buffering mechanisms such as reserve worker force,  
16 ability to compensate for lost or dead workers, things like  
17 that. An effect seen at the colony level is probably realistic.

18  
19 The use of surrogate crops such as Phacelia is  
20 possible the help increase foraging on the crop, or maybe the  
21 crop of interest, and thus insuring exposure. So those are some  
22 of the strengths.

23 Some of weaknesses - study site selection is  
24 probably the single most critical factor in designing a decent  
25 study, competing vegetation can result in low or no exposure to  
26 the target crop and plot size can often be limited when



1 designing these Tier III studies.

2 Again, now plot size will vary with the  
3 attractiveness of the crop in question. So, if you have an  
4 unattractive crop, you may have to have a slightly larger plot  
5 size to ensure adequate exposure.

6 As far as competition, bees will forage on the  
7 most attractive crop within the flight range, and this again can  
8 lead to inadequate exposure to the target crop. So, a knowledge  
9 of the surrounding area is critical and attempts to minimize  
10 competing vegetation is important.

11 The need to treat the control plots to keep  
12 them healthy to some degree may confound the effects that you  
13 may be looking for. So, treating the control plots can be a  
14 major issue, but the lack of treating the control plot may make  
15 it non-comparable because of the health of the plants themselves  
16 may be compromised without treatment.

17 Then lastly, positive control plots are  
18 normally not possible in an environment like this where you  
19 spray a highly toxic compound on a third set of plots to  
20 demonstrate an effect. Those are the things that I listed, the  
21 strengths and weaknesses.

22 **DR. DANIEL SCHLENK:** Thanks. Dr. Ostiguy?

23 **DR. NANCY OSTIGUY:** I concur.

24 **DR. DANIEL SCHLENK:** Dr. Potter?

25 **DR. THOMAS POTTER:** I have one comment. We  
26 may want to make this in several places, is the possible and



1 potential confounding effect of the need to use other  
2 agrochemicals to have agronomic success of your crop may indeed  
3 make it difficult to interpret or evaluate the impacts of the  
4 active ingredient that's the focus of the investigation.

5 **DR. DANIEL SCHLENK:** Okay. Dr. Tarpy?

6 **DR. DAVID TARPY:** I have nothing further to  
7 add.

8 **DR. DANIEL SCHLENK:** Other panel members? Dr.  
9 Ostiguy?

10 **DR. NANCY OSTIGUY:** What I remember as talking  
11 about that, maybe it was with you Jens that we talked about how  
12 if you then applied the material that is applied to the control  
13 crop, you apply it to your treatment crop to attempt at least to  
14 remove that effect from the control crop.

15 **DR. THOMAS POTTER:** You know, absolutely. You  
16 know, in terms of treating your control and hopefully keeping  
17 your materials limited, but even still those other products are  
18 stressors and may ultimately obscure what you're really try to  
19 measure is the effect of the ingredient you're working with.

20 **DR. DANIEL SCHLENK:** That was Dr. Potter. Dr.  
21 James?

22 **DR. ROSALIND JAMES:** Along the lines of those,  
23 sometimes if you don't control pests in your control plot, you  
24 may not have very good flowering and you could have poor control  
25 mortality. Poor growth of your colony and the control  
26 essentially is what your concerned about right, besides the



1 possible interactive effects that you're adding a pest control  
2 measure.

3 The other thing I wanted to add to Dr.  
4 Pettis's list. I hope I didn't miss it. It was the whole study  
5 had to be perhaps repeated in different locations and different  
6 places. Because if you do it one time in a same crop, I mean,  
7 it would be better if it could be repeated in multiple years or  
8 multiple sites and locations. The more times you could repeat  
9 it the better. You didn't really have that, right? Or did I  
10 miss it?

11 **DR. JEFF PETTIS:** That's covered later on. In  
12 trying to say how robust the test would be, it's better to do it  
13 in several geographic areas. We haven't gotten to that.

14 **DR. ROSALIND JAMES:** I apologize. You're  
15 right. That doesn't really address this question.

16 **DR. DANIEL SCHLENK:** Okay. Yes, Dr. Pettis?  
17 Mr. Pistorius?

18 **MR. JENS PISTORIUS:** Just a last comment on  
19 this. I think the uncertainties can be, to a large extent,  
20 avoided. Of course, it should be avoided that if you have a  
21 flowering crop where you do your tests that you apply no other  
22 insecticides. Also with fungicides because then you might run  
23 if that is not the purpose of the study into testing synergisms,  
24 but I think that actually should not be a problem.

25 **DR. DANIEL SCHLENK:** Yes, Dr. Pettis?

26 **DR. JEFF PETTIS:** Just again, back to both



1 those comments. I think the need to have a crop in the control  
2 that's viable, that's producing and flowers and stuff is  
3 important, but you have to balance that. Maybe the best is to  
4 not use a target crop and use something like Phacelia that  
5 wouldn't need those treatments. But again, the best is to  
6 actually apply the formulated material to the crop of interest,  
7 but you have to take measures to make sure the control plots are  
8 not confounding but in good health.

9 **DR. THOMAS POTTER:** Exactly, and I think  
10 those are all things that come into experimental design and I  
11 think are intended to, I guess, provide support for that process  
12 that was described in the White Paper. This is Tom Potter here.

13 **DR. DANIEL SCHLENK:** That was Tom Potter. Any  
14 other strengths or limitations out there? Okay. Let's go ahead  
15 and move onto the next section, question 12.

16 **MR. JOSEPH DECANT:** Joe DeCant, question 12,  
17 subpart (b) - please comment on the proposed modifications to  
18 the field study design elements presented in section 4.3.2 of  
19 the White Paper.

20 **DR. DANIEL SCHLENK:** Dr. Pettis?

21 **DR. JEFF PETTIS:** There will be some  
22 repetition throughout these things, but - so testing the  
23 formulation at the highest label weight is valid. I think  
24 that's a good idea. We agreed that plot location is a critical  
25 step in competing vegetation is an important consideration to  
26 ensure exposure to the target crop. The exposure achieved can



1 accurately simulate the actual use of the pesticide, I think  
2 that's already been stated.

3 The proposed studies are meant to stimulate  
4 worst case scenarios. This is true to the extent that the  
5 pesticide is applied when the highest label rate on the blooming  
6 crop. However, limited plot size may result in a less than  
7 worst case scenario in that these in fact are not going to be  
8 foraging solely on the target plot.

9 Until the pesticide is in actual use and used  
10 on large acreage is you may not see the full extent of the  
11 exposure. So, that's kind of a limitation.

12 I've already pointed out that plot size may  
13 vary depending on the attractiveness of the crop and that less  
14 attractive crops may necessitate having large field sizes. This  
15 is between controlled and treated plots. The exact distance  
16 should may be a minimum of three to six kilometers, but that  
17 distance has to be balanced to get some need to keep those crops  
18 in the same geographic and microclimatic conditions. So, there  
19 is a balance there between the need to keeping them in the same  
20 area and yet keep the distance such that you are not overlapping  
21 the foraging.

22 We agree that balancing or equalizing of bee  
23 colonies is an important step to be taken in advance of placing  
24 the hives in the study site. Otherwise, colony variation  
25 between colonies is too great to see differences. So, colony  
26 equalization prior to placing the colonies into things is a very



1 good step.

2 We agreed that power analysis is a useful tool  
3 to determine the number of colonies needed to produce a  
4 statistically significant result. However, in practice often,  
5 plot size and replication are not such that we can use the power  
6 analysis and those studies aren't often done with high  
7 statistical power. We think the use of a power analysis to at  
8 least guide those studies should be used.

9 The potential endpoints listed are useful to  
10 look at potential adverse effects. I have a couple of specifics  
11 that you list the returning of forager bees. I think you should  
12 be listing something more like the returning of bees over some  
13 time period.

14 I would add that maybe we should add a  
15 consideration of the brood pattern, so you can analyze the  
16 consistency of the brood pattern by measuring 100 cell areas,  
17 and that's not listed on your list. So, there's a way to  
18 measure the consistency or solidness of the brood pattern. We  
19 have shown in other studies that that can be an accurate  
20 predictor of overall colony health.

21 We concur that the duration of the field trial  
22 is going to be variable, and we dictate it by the endpoints of  
23 interest and also by the crop the products are sprayed on.

24 We also concur that opening the hives can  
25 result in increased disturbance from possible introduction of  
26 variation within the results, increased queen loss, things like



1 that increase spikes in adult bee mortality, but the hive  
2 manipulation shouldn't be minimized, but undue concerns about  
3 introducing these artificial effects. So, as long as the  
4 colonies in both plots are treated the same, I think you could  
5 minimize that. I don't think the disturbance factor is so  
6 great. So, hire a good beekeeper basically.

7 And the last comment - most of these field  
8 studies are designed with use of honey bee colonies. So, you  
9 design a large field study and you put honey bee colonies in  
10 there. Certainly, you already have the field design there and  
11 the big problems with these are size of the plot and separation  
12 between control plots and treated plots. The use of things like  
13 Bombus or Osmia added to those plots would not incur a great  
14 deal of cost. Both of those genera fly shorter distances, so  
15 they would be more likely to stay within the field.

16 So most of these Tier III studies are designed  
17 with honey bees in mind with the addition of Bombus or Osmia to  
18 those field studies could be telling in that both they fly  
19 shorter distances, they stay within the treated field and  
20 secondly, they have different life histories. So, you might see  
21 different effects by using two additional species. And that's  
22 all I have.

23 **DR. DANIEL SCHLENK:** Thank you. Dr. Ostiguy?

24  
25 **DR. NANCY OSTIGUY:** I concur.

26 **DR. DANIEL SCHLENK:** Dr. Potter?



1                   **DR. THOMAS POTTER:** I concur with everything  
2 that was said. I just wanted to add that I think there is some  
3 merit in value to possibly incorporating these studies with  
4 terrestrial field dissipation studies or plant residue decay  
5 studies. The more bang you can get for the buck obviously would  
6 be great. So, certainly there is potential for that type of  
7 integration here and certainly something to be contemplated and  
8 encouraged.

9                   **DR. DANIEL SCHLENK:** Dr. Tarpy?

10                  **DR. DAVID TARPY:** This is David Tarpy. I  
11 agree with Jeff's excellent real assessment there and really  
12 have nothing else to add except underscore the utility of the  
13 power analysis and that that's a very useful tool.

14                  **DR. DANIEL SCHLENK:** Other Panel members? Dr.  
15 James?

16                  **DR. ROSALIND JAMES:** I have a few things to  
17 add to the list. In terms of the honey bees, in terms of all  
18 bees, bees are like the rest of us and they are somewhat lazy  
19 and they will only fly as far as they have to generally. So, if  
20 you can keep the - if there is plenty of bee forage available,  
21 they are less likely to fly long distances and leave the  
22 treatment area. So, placing the bees in the middle of the field  
23 and making sure that there's lots of bloom and the design of the  
24 experiment will help. Like a lettuce field, you know, is not  
25 going to have a lot bloom in it.

26                  Then, going back to table 1, which I was in



1 charge of table 1, so I'm going to go back to it. One of the  
2 things that we talked about was that the brood nest size would  
3 be an important measure. So for your list of measures, I would  
4 add brood nest size. That's something relatively easy to  
5 measure.

6 Then, Dr. Pettis brings up a good point about  
7 other bees, and this is a good place now where we can move away  
8 from the honey bee being entirely a surrogate. Since you're  
9 doing a field trial, there are two things to consider. One, you  
10 could have a sample - there's a bee in front of me right now -  
11 you could just go out and do net sampling or pan traps, do some  
12 sort of sampling for what bees are already in the field and  
13 compare between control and treated or before and after.

14 There, you can get your bee diversity measure  
15 in there and hit your third production goal, but also as risk  
16 managers, what is the crop being registered for. If it's being  
17 - the pesticide excuse me - if the pesticide is being registered  
18 for a crop in which something other than honey bees as a primary  
19 pollinator, then maybe the field trial ought to be conducted  
20 with that pollinator instead of honey bees. So, if it's being  
21 registered for alfalfa seed, you ought to be looking at alfalfa  
22 leaf cutting bee. If it's being registered for a tree crop, you  
23 could include something like one of the Osmia bees. So, those  
24 are my additions, thank you.

25 **DR. DANIEL SCHLENK:** Any other input? Yes,  
26 Dr. Hunt?



1                   **DR. GREG HUNT:** I don't know if this was  
2 mentioned, but it's also important to do scouting to see what  
3 the bees are foraging on, to see if they're on nontarget crops  
4 and look at the pollen they're bringing in.

5                   **DR. DANIEL SCHLENK:** Okay. Anyone else?  
6 Okay. Apologies - I forgot to ask you guys on the last  
7 subquestion if you had any comments. If you have any comments  
8 on the last one and this one, any questions of clarification?  
9 Yes, Mr. DeCant?

10                  **MR. JOSEPH DECANT:** Joe DeCant, EFED. Maybe a  
11 clarifying question about the plot sizes - Dr. Pettis, you  
12 mentioned about the different crops given different  
13 attractiveness may require different size of the plots to be  
14 adequate in terms of the field study. Does the panel have any  
15 recommendations or thoughts on minimum plot sizes to be able to  
16 be able bound what we are looking at in terms of the field study  
17 and plot sizes, or a way to calculate how we're going to come up  
18 with the plot sizes or the field study related to the crop?

19                  **DR. JEFF PETTIS:** Jeff Pettis - on plot size,  
20 if you calculate the foraging range of an average colony, even  
21 conservative, and you even to a 1-hectare plot or a 5-hectare  
22 plot, the proportion of the total forage area that they could  
23 visit is really small. So, it's terrible to say, but you know,  
24 larger plots are better and more attractive and reducing  
25 competing vegetation. I would - this is maybe personal and  
26 maybe you can speak to this a bit more. Somewhere, 1 hectare is



1 small, 5 hectares is starting to get, for me, is seemed like you  
2 would be getting in the realm of a reasonable plot size. What,  
3 are you thinking larger? I've seen some of these field studies  
4 where you can get replication. You don't end up having large  
5 fields, I don't know.

6 **DR. DANIEL SCHLENK:** Okay. Who would like to  
7 go first? Dr. James?

8 **DR. ROSALIND JAMES:** I was going to suggest 40  
9 acres for your plot size. I don't think that is ridiculous.  
10 And for seed - so the rules for certified seed and not getting  
11 cross pollination to keep your seed certified, I think, is 1  
12 mile. So, you know, that would be 1 mile between plots.

13 **DR. DANIEL SCHLENK:** Mr. Pistorius?

14 **MR. JENS PISTORIUS:** Well, I think this is one  
15 point where the EPPO needs some modification. Larger field  
16 sizes should be used and described in the old EPPO version, but  
17 I'm quite sure in the next version, there will be an update on  
18 this.

19 I think we have to consider the different  
20 crops. For instance, if we compare maize and winter oilseed rape  
21 - for maize, I would personally go for larger field sizes then  
22 would be necessary for winter oilseed rape. It depends on the  
23 landscape where you do such studies.

24 My personal experience, in former times when I  
25 was young, I conducted such studies a lot. We have a region  
26 where I come from and where I work where you could actually do



1 peer trials, which I think give you the information that you  
2 need with winter oilseed rape of size of one hectare even.

3 I don't know if that is appropriate and I'm  
4 not quite sure the agricultural structure is the same, but we  
5 had a discussion on this too, and it basically depends on the  
6 interpretation of effects and demonstration of exposure. I  
7 think, for instance, if you look at the number of foragers.  
8 Nowadays, we will say okay, maybe this one hectare field size  
9 used to be small for winter oilseed rape in former times.  
10 Nowadays, we say okay, yeah we want to go a little bit larger  
11 because you know in spring, it might be hard to track all the  
12 bees even if you really ensure that in a two or three kilometer  
13 circle, you have no relevant major flowering crops around.

14 It may be different, for instance, if you do  
15 trials with Phacelia because the foraging density is a  
16 completely different one. You get, for instance in our  
17 condition, 5-OH foragers per square meter in winter oilseed rape  
18 and you get about 25 or 30 for Phacelia. So, I think in terms  
19 of these numbers, you may have a certain difference. I think  
20 with one or two hectares, you can start a very bee-attractive  
21 crop and ensuring that there is no cross foraging possible and  
22 landscape which does not provide alternative forage basically.  
23 You will not be able to invite completely, but no major nectar  
24 flow and no major pollen flow. Then you could start to get very  
25 good results with this.

26 When you interpret it as a risk assessor,



1 basically the effects that you get with care, that is a crucial  
2 point because we talked about if you have a controlled level of  
3 mortality of whatever 50. If you get 50 more dead bees, so 100  
4 dead bees after the application, let's assume that it's a  
5 pyrethroid. If you then careful as a risk assessor and say  
6 okay, I've got a concern. Then, it doesn't matter. You know,  
7 if you have five hectares or 50 hectares, the amount of bees  
8 that you will find from your colony is also limited. Because if  
9 you have the exposure maximized and if you basically achieve  
10 that the very large portion of your foragers visit this field  
11 and brings back this contamination - I know this is a crucial  
12 point - then you ensure that those have been well exposed.

13 The number of foragers is limited to some  
14 extent, and you will not get completely different results if you  
15 have good exposure in smaller fields compared to when you have  
16 fields with 50 hectare. It has been the work of Helen Thompson,  
17 the colleague from the U.K. where they have actually addressed  
18 these concerns in studies with the pyrethroid in the U.K. where  
19 they had a field study of I think, one-half or two hectares of  
20 winter oilseed rape. Then they compared it to a landscape  
21 application with 200 hectares. The outcome was not different  
22 from the individual colonies. So, I think, you know, we all  
23 want that the bees are well exposed but maybe depending on the  
24 circumstances, but I think one or two hectares for flowering  
25 crops you're well off. Maybe for pollen and maze, you should  
26 need a little bit larger one.



1                   **DR. DANIEL SCHLENK:** Okay. First Dr. Pettis  
2 and Dr. Ostiguy.

3                   **DR. JEFF PETTIS:** Just to follow up, I would  
4 agree that one hectare maybe on the low end, but for crops such  
5 as Phacelia where you have high visitation rates and in areas  
6 where you don't have competition, they could be adequate. I  
7 would feel more comfortable with five hectare plots for general  
8 crops.

9                   You have to in all cases, trap incoming pollen  
10 and analyze that pollen to guarantee that their visiting the  
11 crop of interest, there is not too much competition, or if there  
12 is competition, it's equal between the control and the treated  
13 plot. So, there are things you can do to guarantee that you're  
14 getting some visitation or reasonable visitation to the plot  
15 size. It's just in practicality that large plots would be  
16 great, but they're just not usually feasible in most settings.  
17 So, five hectares may be - and then maybe down even as low as  
18 one hectare on very attractive crops.

19                  **DR. DANIEL SCHLENK:** Okay. Dr. Ostiguy?

20                  **DR. NANCY OSTIGUY:** This is actually where  
21 Tier III studies are of concern to me because I think that you  
22 get a fair amount of type II error. It helps knowing that the  
23 bees have actually foraged on what you want them to forage on,  
24 but our sizes of plots are too small. I think part of the  
25 difference between what Dr. James said and Dr. Pettis has to do  
26 with differences between the eastern U.S. and the western U.S.



1 Your plot sizes in the west are going to be much larger  
2 generally.

3 I was originally a westerner. That's sort of  
4 my vision also is a bigger area. I recognize though that here  
5 in the east, the size of each plot is smaller, but it does  
6 present a concern to me about type II errors.

7 **DR. DANIEL SCHLENK:** Dr. James.

8 **DR. ROSALIND JAMES:** This is Rosalind James.  
9 This occurred to me too the difference between I think even the  
10 midwest probably, although I have less experience with the  
11 midwest and the west. We are talking about, you know,  
12 three-quarters of the United States. If we throw the midwest  
13 and the west in there together, and especially something like  
14 California agriculture. California is wall-to-wall agriculture,  
15 and there is not a lot of margin places for the bees to hide out  
16 in.

17 So if you have pesticide use, it's going to be  
18 solid and you're talking about cornfields in the midwest, it's  
19 going to be solid. There's not going to be other places for the  
20 bees to gain forage from. Typical irrigation unit in the west  
21 is 40 acres. If you're doing center pivot irrigation, I think  
22 it's 120 acres. So that's one farmer's field that is going to  
23 be very large. Now, so that's one reason why I say 40 acres,  
24 which is what about 12 hectares maybe. It's going to more  
25 because it's three to one. So about 12 hectares would be what  
26 we're talking about.



1 But you could put more than one colony of bees  
2 in the middle of that. You could put four or so there or more.  
3 So, you could put more than one colony in the middle of that and  
4 it's somewhat pseudo-replication. So, your experimental design  
5 is going to be affected by that.

6 **DR. DANIEL SCHLENK:** Dr. Hunt?

7 **DR. GREG HUNT:** This is just a side note.  
8 Regardless of plot size, it may be a good standard practice or a  
9 practice to use sometime to just collect the incoming foragers  
10 and analyze them for pesticides to at least document that they  
11 were exposed. I don't know how good an idea it will give you of  
12 the total level of exposure, but some qualitative documentation.

13 **DR. DANIEL SCHLENK:** Yeah, Dr. Pettis?

14 **DR. JEFF PETTIS:** I think it's a good idea,  
15 Greg. I guess in the studies I've seen that trapping incoming  
16 pollen gives you a really nice measure of the level of exposure,  
17 you know, because you can trace it to the specific crop that  
18 you're growing. I was going to ask you, Dr. James, a question  
19 about - because let's say you can only have one plot of each. I  
20 mean, ideally, you have three or four plots of each type. There  
21 I think I would argue for small plot size. Take your 40 acres  
22 and divide it four ways into four 10 acre plots and get true  
23 plot replication because otherwise, we end up with just putting  
24 10 colonies on this one plot and using the colony as the source  
25 of replication. Ideally, you'd like to use the plot as the  
26 source of replication, but that's not always possible. So, I



1 think that's some of the trade offs.

2 **DR. DANIEL SCHLENK:** Okay. Yeah, Mr.  
3 Pistorius?

4 **MR. JENS PISTORIUS:** I agree, but I think that  
5 actually comes in a later question. It would be desirable to  
6 have those in different environments, then you can actually  
7 cover part E of the question a little bit and do the  
8 representativeness. We do that even in our very tiny Germany.  
9 We ask for northern, eastern and southern or something like that  
10 when we test different environments.

11 **DR. DANIEL SCHLENK:** Okay let's - you still  
12 have another comment? Yeah? Okay. Dr. James?

13 **DR. ROSALIND JAMES:** Watching the clock, he  
14 wants to go home today. But maybe in the west we just have more  
15 land, and a 40 acre plot doesn't seem outrageous to me. I'm not  
16 back down. I think one hectare is way too small. Five - I  
17 don't know, your bees fly a long way.

18 **DR. DANIEL SCHLENK:** So, let me make a  
19 recommendation instead of bantering back and forth. What's in  
20 the written documentation, you can agree to disagree and say  
21 that the positives for the small term plots and then you can  
22 document your justifications for the larger term plots. Yep,  
23 Dr. James?

24 **DR. ROSALIND JAMES:** With my suggestion that  
25 you could sample for bee diversity within the plot, if your plot  
26 is too small, I don't think you can - sampling for bee diversity



1 is not going to work as well because you're going to have the  
2 potential for influx from outside bees into your plot also.  
3 Unless you have immediate acute kill, you may not be able to  
4 tell that there's been an effect on bee diversity. So, if the  
5 plot's too small, that measure may not be effective for you.

6 **DR. DANIEL SCHLENK:** Okay, which one? Dr.  
7 Pettis?

8 **DR. JEFF PETTIS:** Okay. Back to the -- not  
9 dwelling on the larger plot size, but I think the point brought  
10 up by Dr. Pistorius was that if we have a chance of replication  
11 at the plot level, it's probably better across geographic  
12 regions. Then, you gather more information than doing it within  
13 one region. So, testing it in Kansas and Utah and Georgia would  
14 be preferable I think.

15 **DR. ROSALIND JAMES:** The alfalfa seed isn't  
16 grown over all those areas.

17 **DR. DANIEL SCHLENK:** Yes, Mr. Pistorius?

18 **MR. JENS PISTORIUS:** Question to Dr. James.  
19 Did you say sampled bee diversity? Because I think that is a  
20 very complicated measure, because the bee diversity on such a  
21 crop would not be depending on this treatment basically, but how  
22 the population of different bee species was established before.  
23 If you cannot standardize this, then you cannot make basically  
24 an assumption later on how your treatment affected this bee  
25 diversity.

26 **DR. DANIEL SCHLENK:** So, the question, I



1 believe, for clarification was size, right? Is that basically  
2 what the clarification was? I mean, do you want more  
3 information with regard to other endpoints as well?

4 **MR. JOSEPH DECANT:** This is Joe DeCant, EFED.  
5 The main question was about plot size, so I think the panel  
6 touched also on the number of hives per plot. So, we're talking  
7 about, you know, this brings in loading rate as well in terms of  
8 the number of hives per plot. I think that, you know, usually  
9 we're talking about three to four full-size hives at each plot.  
10 So, we're talking about hives of about 50 to 60,000 bees.  
11 Taking that into consideration in terms of the loading rates of  
12 the Panel, and the Panel's recommendations about plot size, if  
13 you also have a recommended loading rate, relating plot size to  
14 the number of hives per plot as well, that would be helpful.

15 **DR. DANIEL SCHLENK:** Okay. That's not  
16 addressed further on in any of the questions? Okay. You might  
17 want to make a statement if you can at the present time so we  
18 can get it on the record.

19 **DR. JEFF PETTIS:** Well, certainly with a small  
20 plot size, you could, by putting large colonies on the small  
21 plots, you could overwhelm the plot and there wouldn't be - but  
22 you still may get some representation, foraging rate and each  
23 colony would get some level of exposure. There is probably a  
24 formula out there that would, depending on the attractiveness of  
25 the crop, you could predict exactly how many colonies. It was  
26 kind of like a pollination, like how many colony spray do you



1 need for almonds, almond crop, things like that.

2 So, there probably are formulas out there.  
3 They're not at the top of my head right now. So, we can address  
4 that a bit in balancing the plot size versus the number of  
5 colonies. Even with and extremely small plot size, you're still  
6 - four colonies per plot would be a minimum because three is the  
7 minimum for any kind of replication.

8 With honey bee colonies, one is going to die  
9 on you automatically. So, you have to start with four. I  
10 prefer to go up from that. But you will recognize that there  
11 could be an overloading effect and try to address that.

12 **DR. DANIEL SCHLENK:** Okay. Yeah, Dr. Klaine?

13  
14 **DR. STEPHEN KLAINE:** If you put multiple hives  
15 on a single plot, isn't that pseudo-replication?

16 **DR. JEFF PETTIS:** It is.

17 **DR. ROSALIND JAMES:** This is Rosalind James.  
18 Or you could consider like a subsample. I mean, you want to put  
19 multiple hives on there because you might lose one for various  
20 reasons.

21 **DR. STEPHEN KLAINE:** Then your replicate is  
22 your plot, not your hive.

23 **DR. DANIEL SCHLENK:** Dr. Pettis?

24 **DR. JEFF PETTIS:** But there is some discussion  
25 about that, because really you want to expose the bees and the  
26 unit that you're measuring is the colony. You're measuring a



1 colony level response. So, in some sense, the colony is the  
2 replicate. But in reality, we would like to have even that  
3 colony replication exposed in multiple plots. That's a better  
4 design.

5 **DR. DANIEL SCHLENK:** So, again, just letter E  
6 I think we talk about study design elements. So, if we can -  
7 yeah. Okay.

8 **DR. ROSALIND JAMES:** I want to answer the  
9 question about how many bees per acre because I think I could  
10 give you a rough guideline of kind of a magic number, 3000 bees  
11 per acre.

12 **DR. DANIEL SCHLENK:** That would be Dr. James.

13 **DR. ROSALIND JAMES:** This is based on what we  
14 do with alfalfa leaf cutting bees, which are slightly smaller  
15 than honey bees and generally the growers put out really about  
16 5000 bees per acre. I'm sorry, 30,000 bees per acre. I will  
17 correct that. From, I think, 30,000 bees per acre is probably a  
18 good rule of thumb. That would be one colony per acre.

19 **DR. DANIEL SCHLENK:** Just to remind the Panel,  
20 we're not here to try to get out quickly, we're just trying to  
21 focus the discussion to the questions that are being asked. So,  
22 that's what my job is, is to try to focus us. It's not to get  
23 us out early and get us home early, just to let you guys know  
24 that. We're just trying to focus the discussion to the question  
25 that's being asked. So, again, Mr. Pistorius you have another  
26 comment?



1                   **MR. JENS PISTORIUS:** Just a question for  
2 clarification basically, how much is 3000 bees per acre?

3                   **DR. ROSALIND JAMES:** 30,000 per acre, so that  
4 would be 2.5 per acre.

5                   **MR. JENS PISTORIUS:** There are 2.5 acres in a  
6 hectare? Okay, thank you. Therefore there is also some  
7 recommendation on the minimal number of bees per square meter  
8 that should be there, and I think for Phacelia before you do  
9 your spray application, for Phacelia it's at least eight and I  
10 will have to look up the number. I think they said two to three  
11 for winter oilseed rape and other crops.

12                   **DR. DANIEL SCHLENK:** Okay. So, it sounds like  
13 several recommendations, which will be placed in the minutes.  
14 Dr. Pettis again, we are relying on you to put those in there  
15 and each panel member to provide their justifications for  
16 whatever numerical value they provide. It sounds like there is  
17 a little bit of disagreement. So, just make sure whatever  
18 number you come across, whether it's hectare size or bee number  
19 that there's a justification for the rest of the panel to weigh  
20 in on. Any other questions of clarification from the Agency for  
21 letter B? Okay. So, let's move onto letter C please.

22                   **MR. JOSEPH DECANT:** Joe DeCant, EFED, question  
23 12, subpart (c) - please comment on factors that should be  
24 considered in evaluating the biological significance of effects  
25 measured in full-field studies in relation to the proposed  
26 assessment endpoints and related protection goals.



1                   **DR. DANIEL SCHLENK:** Dr. Pettis?

2                   **DR. JEFF PETTIS:** Colony growth patterns is  
3 measured by brood production or adult bee population or robust  
4 measures of colony health and should be directly related to the  
5 protection goals or many of the protection goals. Similarly,  
6 things like colony mortality would be obviously biologically  
7 relevant, but you're unlikely to see these adverse direct  
8 effects. The effect on queen survival and queen replacement  
9 would be of biological significance as it has implications for  
10 long-term colony survival and may not be accounted for if test  
11 duration is only of a few weeks or months.

12                   Over winning success may be the real ultimate  
13 test for colony survival, but it's unlikely to be realized  
14 unless the exposure period is closer to the fall and you can  
15 control for factors outside the test period when colonies can  
16 suffer from parasitic mites and things like that.

17                   Adult bee mortality is a good biological  
18 indicator as are foraging rates in adult bee longevity. Each of  
19 these could be good indicators of adverse effects, but effects  
20 could be transitory in nature, and thus extrapolation to the  
21 colony level impacts is not necessarily direct. So, you may  
22 measure those effects, but how they affect total colony survival  
23 is not always one to one.

24                   Taken together any effects noted in the above  
25 endpoints could be used in the risk assessment decision making  
26 even if each endpoint alone was not significant, that is taking



1 the total things measured and looking at those even if they  
2 weren't significant, the weight of impacts could be taken into  
3 consideration as well as Tier I and Tier II testing.

4 Colonies are complex social organisms, and  
5 thus several endpoints measured may be more appropriate than  
6 relying on a single endpoint alone.

7 Then finally, disease in pest levels and  
8 worker longevity, foraging rates and other endpoints that are  
9 measured are less robust, but they should be considered and they  
10 should be actually equalized as far as we can at the beginning  
11 of each study.

12 Any adverse effects noted in things such as  
13 foraging rates or adult bee longevity during the study should be  
14 noted and taken into consideration with any evidence from Tier I  
15 or II testing. So, again, just what effects seen in Tier III  
16 testing should be looked at in context of anything gathered in  
17 Tier I or Tier II. Those were the comments that I had.

18 **DR. DANIEL SCHLENK:** Okay. Dr. Ostiguy?

19 **DR. NANCY OSTIGUY:** I concur.

20 **DR. DANIEL SCHLENK:** Dr. Potter?

21 **DR. THOMAS POTTER:** I concur.

22 **DR. DANIEL SCHLENK:** Dr. Tarpy?

23 **DR. DAVID TARPY:** I agree as well.

24 **DR. DANIEL SCHLENK:** Other Panel members?

25 Okay. Any questions of clarification? All right. We're going  
26 to move on to letter E then. Oh D, sorry.



1                   **MR. JOSEPH DECANT:** Joe DeCant, EFED, question  
2 12, subpart (d) - please comment on factors and methods that  
3 should be considered when extrapolating observed effects at the  
4 colony level in semi-field and field studies to those expected  
5 to occur in the environment, for example spatial and temporal  
6 scale of exposure, hive management practices, presence of  
7 multiple chemical and nonchemical stressors, et cetera.

8                   **DR. DANIEL SCHLENK:** Dr. Pettis?

9                   **DR. JEFF PETTIS:** In Tier II and Tier III  
10 studies, they cannot possibly incorporate all seasonal and  
11 colony management practices that may occur, or changes in  
12 response to the colony due to seasonality changes in colony  
13 growth. But if testing is conducted in the normal growing  
14 season, a certain level of confidence can be had and the  
15 results generated.

16                   So, testing whatever the crop of interest is  
17 would normally be grown and then using colonies during that test  
18 period would be ideal. If you want to do a test and you do it  
19 outside, what would normally be expected as far as seasonality,  
20 then you could have less confidence in the results.

21                   As has been stated earlier, using colonies in  
22 either field or semi-field, they should be uniform in strength  
23 and as disease free as possible at the start of the study to  
24 avoid confounding effects. This is just a personal note - I  
25 would tend to skew towards smaller colonies, although there is  
26 some discussion in the White Paper about larger colonies being



1 preferred. I think well-balanced, small colonies would give you  
2 reliable results and are meaningful biologically.

3 The seasonal nature of colony growth and the  
4 importance of colony preparation for winter, extra stores and  
5 honey and things that gathered, should be taken into  
6 consideration if we're going to try to do a study where we're  
7 going to look at overwintering. So, the exposure period, if  
8 you're going to do an overwintering study, exposure period  
9 should be in the summer or early fall in advance of that to  
10 minimize the amount of time that those colonies are followed  
11 where we have less control of the variables.

12 In Tier III studies, the longer the study  
13 continues or the more variables that will come into play, even  
14 if you've done a good job of equalizing the colonies at the  
15 beginning of the study relative to pest and disease, you're  
16 going to have other outside influences, such as other crops that  
17 they may visit in the interim, additional pesticide exposure,  
18 things that can influence the results of trying to follow these  
19 Tier III studies for longer periods of time.

20 One way to approach this is to take colonies  
21 from the treated plots and the control plots and move them into  
22 a common area that is away from urban or agricultural settings  
23 and try to put them in an as clean and pristine environment as  
24 possible to avoid some of these confounding effectives.

25 Lastly, there could be some consideration of  
26 whether a crop to be tested is a minor or major crop. If it is



1 a major crop, then even the test scenarios under Tier III may  
2 not adequately reflect the ultimate use of the product in such  
3 that these may be exposed more widely. I'm thinking of crops  
4 like soy beans. So, if you test small plot size and you draw a  
5 conclusion and then the products in use are widespread, the  
6 number of acres in things like soy beans may make in reality,  
7 the exposure levels that bees receive in the real world even  
8 greater, and that would be during the test period. So, those  
9 are some of the ideas that we have.

10 **DR. DANIEL SCHLENK:** Dr. Ostiguy?

11 **DR. NANCY OSTIGUY:** I concur, but I'd also  
12 like to add that one of the issues that I have with Tier III  
13 studies is that our goal is to try to approach what are the  
14 ecological conditions that will occur during use of the  
15 material, yet we are at the same time trying to eliminate  
16 confounders. The confounders are part of the reality of the  
17 ecological conditions for how we use honey bees. So, our type  
18 II errors, we're in many ways by our design, increasing the  
19 chance that we're going to find the type II error.

20 On the other hand, we have to control for the  
21 confounders, otherwise you don't have repeatability in your  
22 studies. So, this probably goes in at an evaluation level above  
23 the type II study itself, but there is a very severe limitation  
24 to how good the data actually are for the ecological conditions  
25 we see.

26 **DR. DANIEL SCHLENK:** Thanks. Dr. Potter?



1                   **DR. THOMAS POTTER:** I concur with what Jeff  
2 and Nancy had stated. I have one thing to add. It comes in the  
3 form of a question, and also from long years of experience doing  
4 other types of studies, field studies, and that is the question  
5 of multiple year or studies that have to be conducted over  
6 multiple years to derive some sense of how climatic variables  
7 and other may strongly influence results. So, I do think that's  
8 been addressed here very effectively. Probably an appropriate  
9 topic for discussion.

10                   **DR. DANIEL SCHLENK:** Okay. Dr. Tarpy?

11                   **DR. DAVID TARPY:** My comments have all ready  
12 been made by the others.

13                   **DR. DANIEL SCHLENK:** Okay. Dr. Pettis?

14                   **DR. JEFF PETTIS:** To the question raised by  
15 Dr. Potter, I think it's listed here in the last set of  
16 recommendations. The idea of testing in several geographic  
17 locations may get at that a bit. It certainly doesn't account  
18 for all weather and possibilities, but if you do a test or a  
19 series of tests in several geographic locations, you have a bit  
20 more confidence that the results represented the real world.

21                   **DR. THOMAS POTTER:** I agree, but I don't with  
22 that provisionally. I don't think that simply looking at  
23 different geographic regions addresses the issue of local  
24 climate variability and the potential confounding effect on  
25 results.

26                   **DR. DANIEL SCHLENK:** That was Dr. Potter. Mr.



1 Pistorius?

2 **MR. JENS PISTORIUS:** Dr. Potter also mentioned  
3 the studies conducted over several years and the longer you  
4 conduct such a study, the more you get influence by the  
5 beekeeper and less by the pesticide. What you can do is for the  
6 environmental conditions, what is done in Europe for instance,  
7 if there were no concerns that stopped the registration that we  
8 ask for monitoring in the real world. So, you follow some bee  
9 colonies over a while. Nevertheless, the longer you do it with  
10 exactly the same colonies, treating every colony alike, the more  
11 get an influence of the beekeeping and of individual factors.  
12 Okay sorry. The longer you do it basically with the same  
13 colonies, the more you get an influence of other factors than  
14 pesticides I think.

15 **DR. DANIEL SCHLENK:** Dr. Ostiguy?

16 **DR. NANCY OSTIGUY:** I agree. This is one of  
17 the reasons that I would do a replicate study versus following  
18 the same colonies over years. Certainly, our experience with  
19 the large CAP study, keeping colonies beyond even a year can be  
20 a challenge.

21 **DR. DANIEL SCHLENK:** Dr. Pettis?

22 **DR. JEFF PETTIS:** I will just do a followup.  
23 I think your suggestion of using colonies that are  
24 representative of the real world is a good one. It kind of gets  
25 to Dr. Berenbaum's thing, that in reality, there are sick bees  
26 out there. They're not perfectly healthy. I guess I would



1 argue for reasonably healthy colonies that are uniform in  
2 nature, as disease free as possible, but that's why I would  
3 argue for the small 10 to 20,000 bee colonies that you might be  
4 more likely to see an effect. And you also don't have this  
5 loading issue of putting 60,000 adult bee colonies in there.  
6 So, I would opt for smaller, well balanced colonies that are in  
7 reasonable health.

8 **DR. DANIEL SCHLENK:** Any other comments?

9 Okay. Let me go to the Agency. Any questions of clarification  
10 for letter D? Good. Okay. Let's move onto the last  
11 sub-question there, letter E.

12 **MR. JOSEPH DECANT:** Joe DeCant, EFED, question  
13 12, subpart (e) - a number of study design elements are  
14 discussed in section 4.3.5 of the White Paper. However, even in  
15 the best designed studies, there can be confounding effects  
16 which can limit the utility of these studies in risk assessment.

17 Please comment on factors that should be considered in  
18 determining the utility of field studies for pesticide risk  
19 assessment, including a discussion of the representativeness of  
20 a study for a National Level assessment, that is the pesticide  
21 may be used anywhere in the United States and its territories.

22 **DR. DANIEL SCHLENK:** Dr. Pettis?

23 **DR. JEFF PETTIS:** Again, kind of reiterating  
24 some of the things that were brought out earlier. If the  
25 pesticide is to be used on a widely planted crop common across  
26 the U.S., then testing should be representative and should be



1 done in several environments across the U.S. for it to  
2 representative of a valid study.

3 The ideal test would be using the formulated  
4 product on the crop of interest. In other words, definitely  
5 using it on the exact crop, not on a crop such as Phacelia, and  
6 with the highest level of attraction. So if you had multiple  
7 crops, maybe using it on oilseed rape, whatever the most  
8 attractive crop would be appropriate.

9 If visitation to the crop is confirmed by  
10 analyzing pollen and you still see no adverse effects, then I  
11 think you could have some level of confidence that maybe you're  
12 not producing a type II error that you have good visitation,  
13 crop is sprayed at a high rate, then I think you can have some  
14 level of confidence that the study actually did give you a  
15 reasonable result.

16 The design element, the plot size and plot  
17 separation, we've already gone over. I think they're critical  
18 in designing it. In an appropriate study, the identification of  
19 incoming pollen during the exposure period is probably the  
20 single best measure to give you some level of confidence that in  
21 fact, the exposure has occurred and additional residue analysis  
22 of that pollen can confirm that.

23 I don't know how well that addresses all of  
24 the aspects in E, but maybe they can come out in the discussion.  
25 So, that's what I had.

26 **DR. DANIEL SCHLENK:** Dr. Ostiguy?



1                   **DR. NANCY OSTIGUY:** I concur.

2                   **DR. DANIEL SCHLENK:** Dr. Potter?

3                   **DR. THOMAS POTTER:** I concur. What this  
4 sounds like is an opportunity to do some field work in Hawaii.  
5 So, that sounds good. I do want to add a couple of points here,  
6 and I think that again, it comes back to somewhat of what I said  
7 as a response in the prior question is that we need to very  
8 carefully look at climate and make sure that we're taking  
9 climate into account in terms of its impact on bee populations,  
10 and I'm talking about local climate, you know, in terms of equal  
11 regions within the country as well as variations in climate and  
12 certainly among other things that's year to year, and also of  
13 course issue surrounding climate change.

14                   So, you know, in designing these studies, you  
15 know I guess we need to put our thinking caps on and kind of  
16 work our way through some of these issues related to climate as  
17 well as geography, et cetera, but you know I generally agree  
18 with Jeff and unfortunately don't have the easy answer.

19                   **DR. DANIEL SCHLENK:** Dr. Tarpy?

20                   **DR. DAVID TARPY:** I agree with the comments  
21 that have been said all ready as well, and underscore the  
22 utility of ground truthing and ensuring that the bees are  
23 actually exposed and are foraging on the target crop for a Tier  
24 III study to come back with negative results that does not  
25 demonstrate that the bees were, in fact, foraging on that crop  
26 is not very useful, and that introduces that increased risk of



1 type II errors. So, that's one way to really kind of  
2 standardize across these that the bees are actually doing what  
3 they're thought to be doing.

4 **DR. DANIEL SCHLENK:** Other Panel members? Dr.  
5 James?

6 **DR. ROSALIND JAMES:** I would just add, make  
7 sure you have irrigated and non-irrigated crops because humidity  
8 probably could play a big role and still would call for some  
9 measure of bee biodiversity in the field.

10 **DR. DANIEL SCHLENK:** Dr. Berenbaum?

11 **DR. MAY BERENBAUM:** This may be unanswerable,  
12 but hearing the requirements for plot sizes, you know, they're  
13 sufficiently large that are free of pesticide contamination that  
14 are in reasonably undisturbed areas that are replicated year to  
15 year to accommodate climatic variation. I live in central  
16 Illinois. I don't know that there are any places to put ideal  
17 experimental plot that isn't one way or another compromised.  
18 So, I don't know how to factor this in, but is this a  
19 limitation? I don't know.

20 **DR. DANIEL SCHLENK:** Dr. Pettis?

21 **DR. JEFF PETTIS:** You raise a good point, Dr.  
22 Berenbaum. I think in hearing the German situation, they have  
23 valleys in which there are heavily forested areas. They go in  
24 and plant specific plots up and down those different valleys. I  
25 think in large parts of the U.S. that's impossible and you have  
26 almost at any time the year, you have some competition. One



1 thing I think you can do is look at early spring or summer where  
2 maybe there is less - there's other crops that are maybe there,  
3 but they're not attractive. Your planting them and hopefully if  
4 you're plots are not too far apart, you still got whatever the  
5 other crops of interest - or not crops of interest, the other  
6 competing crops are equal across that. Again, trapping incoming  
7 pollen is a way to measure the amount of visitation to your  
8 target crop, things like that. But there are huge obstacles in  
9 setting up decent plot size and then reducing competing  
10 vegetation.

11 Just one other comment while I've got the mic  
12 - I think it's really important if we're looking at a product  
13 that is going to be used, I will say for a major use versus a  
14 minor crop. If we make an error on a major use and we're going  
15 to license something for use on a major crop, I think there is  
16 more responsibility there than if we make a mistake on a minor  
17 crop use, where the bees may or may not be attractive to the  
18 minor crop but it's not in widespread plantings.

19 Major crops where things are being used, you  
20 know, in like you say the Midwest is a good example. Mistakes  
21 there where we don't adequately test and field test them, then  
22 those should be of concern.

23 **DR. DANIEL SCHLENK:** Mr. Pistorius?

24 **MR. JENS PISTORIUS:** I have to add I don't  
25 live anymore in the hilly areas. It is totally flat where I  
26 live now. Even there, some contract labs are situated and you



1 can still do good tests also in flat landscapes, but we are in  
2 the same situation. It's just a matter of distance between  
3 fields. You may have to choose a little bit larger distances  
4 when it's hilly, but the main thing is the plot isolation.

5 **DR. DANIEL SCHLENK:** Dr. James?

6 **DR ROSALIND JAMES:** I think in the west this  
7 is less of a problem where the majority is agriculture.  
8 Two-thirds of American agricultural products come out of the  
9 state of California. And I have done large scale field  
10 experiments with farmers and you can get replicates in one year.  
11 Also, a lot of its desert, so you only have crop growing where  
12 there is irrigation and you don't have the much competing forage  
13 and forest and things like that, depending on where you do it.  
14 So, maybe a large number of these tests just need to be done in  
15 the west.

16 **DR. DANIEL SCHLENK:** Any other comments?

17 Okay. We go to the Agency. Mr. DeCant, you have any questions  
18 of clarification?

19 **MR. JOSEPH DECANT:** Joe DeCant, EFED. I have  
20 one question of clarification. So, the Panel mentioned about  
21 the field studies and the design of the field studies and making  
22 sure that we have adequate plot sizes and looking at the  
23 chemicals under good agricultural practices, but at the same  
24 time, the Panel is recommending looking at foraging. So, part  
25 of this speaks to the study design and even if we have adequate  
26 study design, then we also need to ensure that the bees are



1 foraging. So, my question to the panel is do you have any  
2 recommendations or thought on what would be considered an  
3 adequate level of foraging, either in the case of if we identify  
4 a difference between the control and the treatment in terms of  
5 where they're foraging or just an overall foraging and what we  
6 would consider as adequate.

7 **DR. DANIEL SCHLENK:** Dr. Pettis?

8 **DR. JEFF PETTIS:** I'll take a stab at that.

9 Knowing that you can't control - I mean, you've got a wide array  
10 of options out there to forage on, just as a ballpark figure, if  
11 50 percent of the pollen was coming back from the target crop, I  
12 would think that would be good exposure. Anything above that is  
13 even better. And then, are you interested in foraging rate?  
14 Are you interested in something about foraging rate - I think  
15 just determining that they are foraging on the crop of interest  
16 and using the incoming pollen and the indentifying of that  
17 pollen such that you have some assurance that a reasonable  
18 amount of the exposure has been undertaken. For lack of a  
19 better - off the top of my head, 50 percent of the pollen, you  
20 know you're getting 50 percent of their diet from that target  
21 crop. With Dr. James's 48 acres, that would be easy to do,  
22 right? Acres, yeah. I don't think you want approach 100  
23 percent. You're just not going to get there, but anything 50  
24 percent or greater of the incoming pollen would indicate a  
25 reasonable amount of foraging on the target crop.

26 **DR. DANIEL SCHLENK:** Let me just go through



1 their main ones. Dr. Ostiguy, do you have a number, percentage?

2  
3 **DR. NANCY OSTIGUY:** No, I don't think there  
4 really - I don't know of any data that actually gives us that  
5 information. I'm just still back to my problem with type II  
6 errors.

7 **DR. DANIEL SCHLENK:** Okay. Dr. Potter?

8 **DR. THOMAS POTTER:** Getting back to what Dr.  
9 Berenbaum said about Illinois, certainly that's the case in some  
10 other parts of the country where you have the landscape  
11 literally heavily disturbed, but also covered with vast wasps of  
12 crops of interest. So, in the context of these studies, I'm  
13 trying to get my head around how you do pollen counts or pollen  
14 typing and are assured that that's indeed from the pesticide  
15 treated crop. So, I guess I'm asking a question here and maybe  
16 Dr. Pettis has some thoughts on that.

17 **DR. DANIEL SCHLENK:** Dr. Pettis?

18 **DR. JEFF PETTIS:** Obviously, you couldn't if  
19 you were treating canola. You couldn't have competing canola  
20 fields because they would be confounding. So, whatever your  
21 target crop that you're planting would need to be unique. So,  
22 you would have to be able to identify, and then you do analyze  
23 that 50 percent of the incoming pollen is from canola, and you  
24 analyze the pesticide level of that versus control to see if  
25 there's been crossing between the plots. It's fairly straight  
26 forward, and I agree that Midwest, it would be very difficult.



1 I'm thinking desert, Southwest and these nice big circles. When  
2 you fly out west and you see these nice big circles, those are  
3 idea places of to do that.

4 **DR. THOMAS POTTER:** Yeah, I brought that up  
5 because I don't - that is what I wanted to capture in the sense,  
6 in our notes here. This is Tom Potter.

7 **DR. DANIEL SCHLENK:** Thanks. Dr. Tarpy?

8 **DR. DAVID TARPY:** This is Dave Tarpy. You  
9 want a percentage from me or just to follow up on that?

10 **DR. DANIEL SCHLENK:** Well, I think the EPA is  
11 wanting a measure of foraging success.

12 **DR. DAVID TARPY:** So, to Dr. Pettis's point  
13 then on that, I think 50 percent is probably a good target. I  
14 mean in some cases, that might be high. I don't know if there  
15 is a magic number as far as percentage of returning pollen  
16 foragers with target crop pollen on their corbiculae, right?  
17 Because there are a lot of target crops where they are not  
18 pollen bearing at all and they are not going to be collecting  
19 any pollen, right? So, that may be difficult.

20 What I was referring to as far as ground  
21 truthing was actually doing things like transect walks or what  
22 Dr. James was saying of the pan traps and actually collecting  
23 bees in those traps to verify that bees are actually in the  
24 field of interest. Actually, doing transects up and down the  
25 rows of that particular crop and visibly verifying that honey  
26 bees and other are foraging on those flowers. Therefore,



1 putatively coming into contact with and being exposed with the  
2 chemical in question.

3 There are many ways to get to that, and it's  
4 probably crop dependent as to the best measure of verifying that  
5 honey bees are actually in the target crop.

6 **DR. DANIEL SCHLENK:** Okay. Other panel  
7 members? Dr. Hunt?

8 **DR. GREG HUNT:** Yeah, these studies are so  
9 hard to set up. I don't know the answer to the question, other  
10 than to say that you ground truth, you measure the pollen,  
11 proportion, whatever it is and report it, and what was the other  
12 thing I was going to say - oh yeah, foraging rate, it depends on  
13 if what you're applying is expected to be like a foliar  
14 application and it's expected to be a burst of time point. Then  
15 you would want to take the foraging rate that day.

16 **DR. DANIEL SCHLENK:** Okay. Any other  
17 comments? Oh sorry, Mr. Pistorius?

18 **MR. JENS PISTORIUS:** Well, depending on the  
19 crop, it's very different. For instance, Phacelia, you can do  
20 very easily an optical differentiation because it's a purple  
21 pollen. Almost no other crop has a purple pollen. You can  
22 easily count by the numbers of pollen grain for that color.  
23 That is Phacelia pollen, if you have ensured that there are no  
24 other Phacelia crops around, which I assume no. It may be more  
25 difficult with, for instance, canola, because at that time of  
26 blooming probably the same year, you get a few other flowers or



1 some other flowers in the surrounding, which also have a  
2 yellowish color. So, you have to do pollen analysis.

3 Then what I still think is a very good measure  
4 is foraging, the number of bees foraging on that specific crop.  
5 For us for our conditions, we have by experience, we have  
6 minimum numbers. For instance, for winter oilseed rape, if we  
7 know that if it's below two or three foragers per square meter,  
8 it is not valid because also depends on the bee size. For  
9 Phacelia, you can say that normal is at least 12 to 25. I think  
10 those numbers may also - I mean we would have to look at other  
11 crops to find certain numbers. I mean for a tree, it is  
12 probably more difficult to estimate, but our possible estimates  
13 to ensure that the bees have actively been foraging on the crop.

14  
15 **DR. DANIEL SCHLENK:** Yes, Dr. Fefferman?

16 **DR. NINA FEFFERMAN:** Sorry, I just have a  
17 question and listening to all this. Hopefully, this question is  
18 really stupid, but it gets back to something Dr. Berenbaum  
19 mentioned about choice. I'm wondering if we separated the  
20 exposure route from actual ingestion versus contact if we should  
21 worry at all about landing rates on crops that are then rejected  
22 as forage, if that's a contact exposure without the concomitant  
23 ingestion exposure.

24 **DR. DANIEL SCHLENK:** Again, I think we're kind  
25 of trying to answer a design question here with this. Okay.  
26 Anybody want to chime in? Yeah, Dr. Hunt?



1                   **DR. GREG HUNT:** Well, if I understand the  
2 question, you're talking about bees visiting a flower and then  
3 rejecting it. Once they reject that flower, they've all ready  
4 landed on it. I think you'll get this from the scouting. You  
5 know, if they continue to reject, they will continue visiting  
6 flowers of the same kind. But pretty soon, they're going to  
7 look for something better.

8                   **DR. NINA FEFFERMAN:** This is Nina Fefferman  
9 again. This is basically my thought in response to Dr. Tarpy's  
10 idea of watching who's flying where and having that mismatch  
11 with what they're doing. So yeah, I agree over time if they're  
12 going back and communicating that it wasn't a good source and  
13 they have to go elsewhere, yeah they just won't keep visit the  
14 area. But what you're locally doing is walking a transect and  
15 just looking, let's say early in the morning for where scout  
16 foraging is going. That might be a real disconnect in what's  
17 happening.

18                   **DR. DANIEL SCHLENK:** Okay. Does that answer  
19 your question in regards to foraging? Any other questions of  
20 clarification? Okay. Let's go ahead and move on to 13.

21                   **DR. THOMAS STEEGER:** This is Tom Steeger,  
22 question 13A please comment on the use of data on individual  
23 bees to transition to higher tier studies given that the Tier I  
24 studies focus on survival as the primary measurement endpoint  
25 although additional endpoints may be forthcoming as test designs  
26 continue to develop.



1                   **DR. DANIEL SCHLENK:** Our lead discussant is  
2 Dr. Tarpy.

3                   **DR. DAVID TARPY:** This is Dave Tarpy, NC  
4 State. So, many of these points or at least for this  
5 sub-question, I think have been articulated all ready. For the  
6 sake of time and just to not have to recapitulate them all over  
7 again, we'll just refer to those previous questions. So, this  
8 is starting to funnel up a lot of our discussions from yesterday  
9 and today on this and starting to distill them.

10                   In general, much of those discussions, it  
11 seems pretty clear that there isn't this perfect proxy of Tier I  
12 and lower tiers that really capture a lot of the complexity of  
13 the upper tiers. This is especially true in social systems like  
14 in honey bees. So, additional care needs to be taken in trying  
15 to equivocate these different tiers.

16                   So, because of that, I think a lot of what  
17 we've been talking about in question 8, question 11 in  
18 particular about blurring the lines between the Tier I, II and  
19 III in using groups of individuals rather than individual bees,  
20 not solitary bees. But to incorporate bioassays that test the  
21 social unit will go a real long way to addressing some of this.  
22 Certainly not completely, but it really does help to capture the  
23 biological reality of looking at colony and group phenotype  
24 rather than just individual phenotype.

25                   With of course the exception of queens and  
26 drones, where looking at individual phenotypes on their



1 reproductive potential is something that would be obviously  
2 worthwhile. We also agree as the discussants that mathematical  
3 modeling will be able to help hopefully tie a lot of these types  
4 of aspects together, particularly if the same phenotypes are  
5 measured at these different levels of reductionistic to larger  
6 scale types of experimental designs. So if the same phenotypes  
7 can be measured at those different increasing levels being able  
8 to kind of verify them across those different levels of  
9 biological organization would be very powerful means of  
10 verifying that and using mathematical modeling to have  
11 predictions from one level to another.

12 We also, and Dr. Berenbaum can extrapolate on  
13 this if she wants, but that data on the individual pupae can be  
14 very informative at lower levels to higher levels. That doing  
15 Tier I bioassays on contact exposure on individual pupae, that  
16 is likely to have very strong penetrants to colony-level  
17 phenotype compared to some of the other more nebulous ones like  
18 we've been talking about of subtle behavioral differences, which  
19 again can have very important biological relevance, but much  
20 more difficult to capture and to translate from one level to  
21 another.

22 **DR. DANIEL SCHLENK:** Dr. Berenbaum?

23 **DR. MAY BERENBAUM:** What I was going to say  
24 was said, so I concur.

25 **DR. DANIEL SCHLENK:** Great. Dr. Pettis?

26 **DR. JEFF PETTIS:** I concur.



1                   **DR. DANIEL SCHLENK:** Other Panel members?  
2       Must be getting late in the day. Oh, sorry, Mr. Pistorius?

3                   **MR. JENS PISTORIUS:** Well, I just want to make  
4       a comment, for instance, on the IGRs. I may have made this  
5       comment all ready. I think for IGRs for instance, that the  
6       current Tier I studies as they are designed, also taking into  
7       account that there's a seven-day Aupinel method available soon,  
8       we hope. Unless the Aupinel method is soon also extended to the  
9       pupation and the hatching phase, that just looking at the  
10      individual mortality data of adult bees as measured by acute  
11      oral or contact toxicity, we would miss the effects of IGRs.

12                   In principle, I agree that this data on the  
13      individual bees, the transition to higher tier tests is a  
14      reasonable approach given some limitations, but I still think  
15      that there must be some flexibility for the risk assessor to  
16      identify, for instance, like the IGRs, the need for other  
17      information like the mode of action go to higher tier tests  
18      immediately or in addition. So, saying that for the risk  
19      assessor, there is a need that we don't have a calculation  
20      somewhere. This is limiting the option to go into higher tiers  
21      because currently there would be, or if you say okay, no trigger  
22      bridge by what we know. Some things could be missed.

23                   **DR. DANIEL SCHLENK:** So, it sounds like you  
24      would like at qualitative component to that sort of decision  
25      process there. Any other comments on letter A? Okay. Let's  
26      move forward to letter B.



1                   **DR. THOMAS STEEGER:** This is Tom Steeger,  
2 question 13B - please comment on the derivation of the Level of  
3 Concern, that is the LOC of 0.4 and the extent to which is  
4 serves as an appropriate screen to transition to higher tiers of  
5 testing and refinement.

6                   **DR. DANIEL SCHLENK:** Dr. Tarpy?

7                   **DR. DAVID TARPY:** So this is Dave Tarpy. The  
8 discussants on this particular sub-question would also like to  
9 hear from many of the toxicologists on the panel to provide  
10 their insights since none of us really are toxicologists, but it  
11 seems to be that our take on this in distilling a lot of the  
12 other discussions, that level of concern as derived at kind of  
13 lower tier more abductionistic levels are a good empirical way  
14 to arrive at those kind of baseline levels that need to be  
15 vetted and verified at upper levels. Those are obviously a very  
16 good starting point.

17                   But because of the transient property from one  
18 level to another is not 100 percent obviously, that I think that  
19 that would provide a good starting point, but it's not  
20 necessarily going to be exactly the same as one increases in  
21 biological complexity and in environmental variability. So, it  
22 seems an appropriate starting point, but that it should be  
23 modulated depending on the different tiers in which they're  
24 being tested.

25                   **DR. DANIEL SCHLENK:** Okay. Dr. Berenbaum?

26                   **DR. MAY BERENBAUM:** I just like to repeat the



1 call for input from our toxicologist colleagues who have  
2 examined these kinds of estimates in much broader context than  
3 we have.

4 **DR. DANIEL SCHLENK:** Dr. Pettis, let me guess  
5 what you're going to say.

6 **DR. JEFF PETTIS:** I'm gonna say that our level  
7 of concern among the three of us was raised that we were looking  
8 for help basically.

9 **DR. DANIEL SCHLENK:** Okay. With that, anybody  
10 want to jump in? Dr. Klaine, not saying any names.

11 **DR. STEPHEN KLAINE:** You know, this thing is  
12 really confounded by the fact that as you move from one level to  
13 another. You've got this issue of single organisms moving to  
14 more complex systems. And I'm really not sure if we're able to  
15 quantify the level of concern that would be the most useful as  
16 you move into these higher tiers.

17 **DR. THOMAS STEEGER:** Could I make a point of  
18 clarification?

19 **DR. DANIEL SCHLENK:** Sure, Dr. Steeger.

20 **DR. THOMAS STEEGER:** The level of concern is  
21 applied to the Tier I laboratory-based studies only. The higher  
22 tiers studies that are at the colony level do not have a level  
23 of concern that's applied to them.

24 **DR. DANIEL SCHLENK:** Thanks for that  
25 clarification. This is just for the Tier I.

26 **DR. STEPHEN KLAINE:** All right. Okay. I



1 misread the question.

2 **DR. DANIEL SCHLENK:** Are you done? No? Are  
3 you thinking?

4 **DR. STEPHEN KLAINE:** For the moment.

5 **DR. DANIEL SCHLENK:** Okay. Dr. Hunt?

6 **DR. GREG HUNT:** Well, my understanding is this  
7 LOC 0.4 corresponds to about 10 percent mortality. Given that a  
8 lot of the suggestions are to follow the larvae to pupation and  
9 to have longer observation of adults in survival analysis, I  
10 don't see how this particular number applies. But probably some  
11 number could be developed or some qualitative, you know, in  
12 terms of development maybe, there could be a number developed  
13 there as well. But, anything that is enough standard deviations  
14 from the norms.

15 **DR. DANIEL SCHLENK:** Mr. Pistorius?

16 **MR. JENS PISTORIUS:** With limitations of not  
17 knowing what exactly which substances, which products exactly  
18 were behind the data, that table that Dr. Steeger showed the  
19 other day on how many substances would actually pass the first  
20 trigger. Given that there is a need in my opinion for tiered  
21 approach in risk assessment and also keeping into account that  
22 we might miss a very small number, I think really a very small  
23 number of active with such a tiered approach, I have the  
24 impression that this level of concern is a conservative measure  
25 and an appropriate screen considering the comparison with the HQ  
26 data, which has been for a long time used in Europe and very



1 well information we have on incidents in different countries.  
2 The clearly improved conservatism compared to this approach, I  
3 don't remember which presentation it was but, in one  
4 presentation it was said that this approach is about a factor  
5 3.6 more conservative than the approach that we had. So that  
6 might be a German interpersonal perspective.

7 **DR. DANIEL SCHLENK:** Thanks. Dr. Klaine?

8 **DR. STEPHEN KLAINE:** Yeah, I agree. It's very  
9 conservative. My hesitation before really comes from trying to  
10 get my head around the fact that when we do this with other  
11 species, when you have an acute number, you get some reflection  
12 about the impact on the population. In this case, we did it  
13 with single organisms, and we really don't know how that  
14 reflects that actual health of the hive or the ultimate  
15 population of bees. So, I agree it's conservative. I'm  
16 actually bordering on the fact that it might be too  
17 conservative. I'm not sure how to quantify that.

18 **DR. DANIEL SCHLENK:** Anyone else want to weigh  
19 in on that? For what it's worth, I think it's conservative in  
20 terms of you know, what it's trying to do and the function that  
21 it's trying to serve in a Tier I screen that seems to have very  
22 limited error with regard to false negatives, which is what  
23 you're trying to do, right? Yes?

24 **DR. STEPHEN KLAINE:** Yeah, and where I  
25 hesitate is what is the right number.

26 **DR. DANIEL SCHLENK:** Isn't it 42? Yes? Dr.



1     Pettis?

2                    **DR. JEFF PETTIS:** I guess just a question to  
3     EPA and Dr. Hunt touched on this. This value roughly relates to  
4     10 percent mortality, and that would be above whatever the  
5     control mortality is. So, if you had control mortality of 10  
6     percent, then you would have to see 20 percent mortality in  
7     another group. Is that correct?

8                    **DR. THOMAS SEEGER:** This is Tom Seeger.  
9     Essentially it's taking that LD50 value. The slope that is  
10    calculated off of all of the acute toxicity data that we have  
11    for oral and contact toxicity and estimating what would the  
12    value have to be to back it down to control mortality. So,  
13    you're essentially bringing it down to a NOAEC that would be -  
14    well you're not able to distinguish between the treated and the  
15    control value. So, the chronic LOC is set to 1, because the  
16    NOAEC is compared to the exposure value and the acute value  
17    because we're backing that LD50 back down to what we think is  
18    background mortality, acute mortality is set to 10 percent.

19                   **DR. DANIEL SCHLENK:** Dr. Berenbaum?

20                   **DR. MAY BERENBAUM:** Point of clarification  
21    that you say all available acute mortality data, toxicity data,  
22    are those from forager or workers, right? Are there any larval  
23    data that is factored in?

24                   **DR. THOMAS STEEGER:** These data were based on  
25    adult - young worker data, that's correct.

26                   **DR. DANIEL SCHLENK:** So, do we have a



1 consensus that 0.4 is valid? That's what the question is  
2 asking, right? Yes? If you don't then you need to come up with  
3 another number, I guess. That's the question. Yeah, Dr.  
4 Klaine?

5 **DR. STEPHEN KLAINE:** Steve Klaine. You know,  
6 the goal is to be conservative and to not make errors. So, you  
7 know if we're gonna go in that direction, then this is in fact  
8 conservative. I think that as more data become available in  
9 terms of the relationship between individuals and hive health, I  
10 think that you may want to hold out the opportunity to change  
11 that.

12 **DR. DANIEL SCHLENK:** Yeah, what I would say  
13 too is particularly if you're going to change the assay to the  
14 other assays that have been discussed, that number may actually  
15 change as well if we go with some of the alternative assays that  
16 have been proposed, at least durations. Yeah, Dr. Fefferman?

17 **DR. NINA FEFFERMAN:** So, this is actually a  
18 strange context for me to propose this, but this is actually one  
19 of the context in which I would propose a sensitivity analysis  
20 from the end, not of the risk assessment, but for the risk  
21 manager. Because the only danger I see in being too  
22 conservative is that if you accidentally make it so conservative  
23 that almost every viable benefit already puts you over the  
24 threshold, then it makes it a yes or a no, do we want the  
25 benefit at all question.

26 So, if there is a way to do a sensitivity on



1 the relative benefits from the management perspective in  
2 interpreting the outcome of the risk assessment based on the  
3 sensitivity of the cutoff for how conservative that should be,  
4 there is a utility of not being so conservative that the answer  
5 is anything you do is equally bad, so maximize your gains if  
6 you're going to do anything because that can push - am I being  
7 clear enough? Okay. Cool.

8 **DR. DANIEL SCHLENK:** Did you get that Dr.  
9 Tarpy? Any questions of clarification? Yeah, we're good?  
10 Okay. We're going to go forward to letter C then.

11 **DR. THOMAS STEEGER:** This is Tom Steeger,  
12 question 13C - please comment of the quantitative aspect of the  
13 screening-level Tier I assessment and the use of Tier II and  
14 Tier III whole-hive studies to qualitatively characterize risk.

15 **DR. DANIEL SCHLENK:** Dr. Tarpy?

16 **Dr. DAVID TARPY:** This is Dave Tarpy. I think  
17 our comment on the answer to this one is because it is very  
18 similar to the others about extrapolating from one level to the  
19 others is a little more difficult in light of the social life  
20 history of honey bees. So, this pretty much the same caveat is  
21 given as to the previous question.

22 **DR. DANIEL SCHLENK:** Dr. Berenbaum?

23 **DR. MAY BERENBAUM:** I concur.

24 **DR. DANIEL SCHLENK:** Dr. Pettis?

25 **DR. JEFF PETTIS:** I concur.

26 **DR. DANIEL SCHLENK:** Any other panel members?



1 So what it sounds like is this has been addressed in earlier  
2 questions. Are you guys okay with that? Yeah? Okay. Letter  
3 D.

4 **DR. THOMAS STEEGER:** This is Tom Steeger,  
5 question 13D - please comment on the assumption that the  
6 effects on individual bees measured in laboratory studies must  
7 be considered in the context of whole colony studies conducted  
8 under semi-field and full-field conditions.

9 **DR. DANIEL SCHLENK:** Dr. Tarpy.

10 **DR. DAVID TARPY:** This is Dave Tarpy. Again,  
11 as has been covered in many of the questions. I think it has  
12 been pretty well flushed out, but the assumption that  
13 individuals serve as a valuable proxy for the whole group is  
14 pretty much nullified and doesn't exist, which is why we spend  
15 so much time emphasizing alternative means of testing groups in  
16 vitro and in vivo to try to more realistically approximate in  
17 vivo colony dynamics.

18 So, there should be real degrees of care to  
19 make broad-sweeping inferences of data on at the individual  
20 honey bee level to full-field colony phenotypes. So, by being  
21 able to assess at Tier I with social groups and kind of more  
22 biologically relevant bioassays that will go a long way to  
23 alleviate this particular question.

24 **DR. DANIEL SCHLENK:** Dr. Berenbaum?

25 **DR. MAY BERENBAUM:** I concur.

26 **DR. DANIEL SCHLENK:** Dr. Pettis?



1                   **DR. JEFF PETTIS:** I have just a bit to add.

2       Just in looking at effects that may be realized in Tier I, they  
3       may help guide you and then what you might look for in full  
4       colony or semi-field or field studies, but there is a caution.  
5       I think Dave has already touched on it that an effect that you  
6       might see in a cage with individual bees, there are so many  
7       compensation mechanism of grooming and prophylaxis and things  
8       that happy at the colony level.

9                   That effect may totally disappear and you may  
10      be looking for other effects that may happen at the colony  
11      level. Food transfer may magnify affects towards the queen,  
12      things like that. So, I think it's important to look at the  
13      effect that Tier I and move forward. But the social dynamics  
14      and colony interactions can really - it's really hard to predict  
15      how those individual measures might be realized in the colony.  
16      So again, kind of reiterating what is being said.

17                  **DR. DANIEL SCHLENK:** Other panel members?

18      Okay. We go back to the Agency. Any questions of  
19      clarification?

20                  **DR. THOMAS STEEGER:** Point of clarification.

21      In the absence of those other socially proper studies, the  
22      current guidelines that exist serving as the basis for Tier I  
23      studies and using them in a screen, does the Panel concur or can  
24      you comment on the use of the currently available guidelines.

25                  **DR. DANIEL SCHLENK:** Dr. Tarpy?

26                  **DR. DAVID TARPY:** So, this is Dave Tarpy. I



1 guess that they are informative, but not as informative as they  
2 could be because of those complexities. So I think, and the  
3 Panel can agree or disagree, but that's why we've spent so much  
4 time urging the development of more biologically relevant in  
5 vitro bioassays that would kind of harness that a bit better. I  
6 mean obviously, some informed information is better than no  
7 information, but I think that because of this disconnect of the  
8 individual versus group level that those data derive at the  
9 individual level ought to be interpreted with much greater  
10 caution.

11 **DR. DANIEL SCHLENK:** Dr. Berenbaum?

12 **DR. MAY BERENBAUM:** One additional benefit of  
13 using the existing studies as approaches is that it would allow  
14 harmonization of international efforts and an acquisition of a  
15 greater data base to use in trying to make that connection. But  
16 I just want to echo the call for reexamining assay design to be  
17 more reflective of biological reality.

18 **DR. DANIEL SCHLENK:** Dr. Ostiguy?

19 **DR. NANCY OSTIGUY:** I want to actually  
20 strongly support the use of Tier I data this point, because we  
21 don't have anything else to work with. It is much better than  
22 nothing. Actually, so much better than nothing that it's almost  
23 no comparison, which doesn't negate the fact that there might be  
24 better ways to assess things. I do think that they will provide  
25 us with a sufficiently sensitive set of criteria for making  
26 decisions that we should not commit too many type II errors.



1                   **DR. DANIEL SCHLENK:** Okay. Mr. Pistorius?

2                   **MR. PISTORIUS:** Yes, I think that the current  
3 guidelines are very suitable. We know that there are a few  
4 guidelines or there is especially one guideline, which has to be  
5 further developed or established at all at the Tier I level.  
6 This is yet to be done. I would say as a general remark that  
7 some of the existing guidelines, also the OECD 75 and EPPO 170  
8 need a little bit of updating. But I think those were the  
9 points that were discussed in our discussions here already, and  
10 they are minor points, which do not criticize the whole approach  
11 that is mentioned in the guidelines.

12                   **DR. DANIEL SCHLENK:** Dr. James, did you have  
13 you hand up earlier? No? Yes?

14                   **DR. ROSALIND JAMES:** I don't like the current  
15 guidelines. I think they're - we're having problems with bees.  
16 They are not a very good assessment of what our effects are  
17 going to be. So, I guess I don't know that they are much better  
18 than nothing. We're talking about the 48 hour contact toxicity  
19 on adult tests, right? When you say current guidelines? At  
20 Tier I. They're recommended in the White Paper or the current  
21 guidelines, I'm not sure what you're asking I guess.

22                   **DR. THOMAS STEEGER:** This is Tom Steeger,  
23 point of clarification. Two points of clarification. We are  
24 talking about now actually quantifying risk using a formal  
25 process, which has not been done before. We are also talking  
26 about modifications to the testing process that will give us a



1 much better understanding of not just adult toxicity, but larval  
2 toxicity and a path forward to where there are potential risks  
3 that are identified using these screening level tests to  
4 transitioning the higher tiered tests so would better enable us  
5 to understand whole colony effects.

6 **DR. ROSALIND JAMES:** This is Rosalind. And  
7 when you said current guidelines, that's what you're referring  
8 to?

9 **DR. THOMAS STEEGER:** This is tom Steeger. I'm  
10 discussing what has been laid out in the White Paper.

11 **DR. ROSALIND JAMES:** Okay.

12 **DR. DANIEL SCHLENK:** Okay. Dr. Fefferman?

13 **DR. NINA FEFFERMAN:** So I think the Tier I  
14 studies for individual level are really good for the individual  
15 effects. Where I find the disconnect for the context of the  
16 question of scaling those up is actually propagates back down,  
17 and I don't remember seeing quantitative ways to characterize  
18 the sublethal effects that sufficiently trigger Tier I response  
19 to go to Tier II. And I don't know if that's because I'm just  
20 not remembering it or because it's not there. I know there was  
21 discussion of how to try to measure some sublethal effects, but  
22 I don't remember seeing how that gets interpreted in the  
23 conceptual flow model for then going from okay, we've finished  
24 our Tier I study, we saw the sublethal effects. That means we  
25 now really do need to go to Tier II. That's the part where I  
26 start not knowing how to get my mind around if this working



1 well.

2 **DR. DANIEL SCHLENK:** Any other comments? Dr.  
3 Hunt?

4 **DR. GREG HUNT:** I think the guidelines  
5 proposed in the White Paper are a good advance - a good  
6 improvement. But I don't think there was proposed the use of  
7 sublethal effects. In Tier I, it was proposed as something that  
8 might be useful in the future. I think that what the panel is  
9 saying is that there could be improvements in sublethal effects  
10 included for example, following the growth of the larva to  
11 pupation and looking at biological endpoints and things of that  
12 sort. So I think that what's proposed, if I'm understanding  
13 correctly is good, but we're suggesting there could be some easy  
14 improvements. Maybe not so easy.

15 **DR. DANIEL SCHLENK:** Any other comments? Dr.  
16 Klaine?

17 **DR. STEPHEN KLAINE:** Yeah, I just want to  
18 weigh in on this. From the panel discussions and all the  
19 discussions we've had, it just at that level, it appears that  
20 mortality is just the wrong endpoint and it's difficult to  
21 interpret to move forward with that. I think that's why  
22 everyone is calling for sublethal endpoints. So that the issue  
23 with behavioral endpoint or other endpoints as they are  
24 developed would better able this movement from single organisms  
25 in Tier I up to the semi-field and field studies.

26 **DR. THOMAS STEEGER:** Point of clarification?



1                   **DR. DANIEL SCHLENK:**    Sure.

2                   **DR. THOMAS STEEGER:**   For the - in the White  
3 Paper, it describes that the sublethal effects at this time  
4 because of the absence of appropriate linkages to assessment  
5 endpoints, while they would not be used quantitatively, they  
6 would be used qualitatively and that they would be considered  
7 with the other lines of evidence available to determined whether  
8 higher tier testing would be required. So, the fact that the  
9 current battery of Tier I studies tend to focus on mortality as  
10 the primary effect. There is the opportunity that additional  
11 endpoints may be reported, but we're not confined to only  
12 considering the mortality endpoint. We do leave open the option  
13 to qualitatively consider whether the sublethal effects are  
14 sufficiently compelling to warrant with other lines of evidence  
15 transitioning to higher tier testing.

16                  **DR. DANIEL SCHLENK:**   I believe that is going  
17 to be the next sub-question, right? Yes. Dr. Bradbury, did you  
18 have something you want to say? No, he just got - okay. Great.  
19 Well with that, are you okay with the answer that we received  
20 for this sub-question? So, let's call it quits for now. I'm  
21 sensing a little bit of brain drain here this afternoon. So,  
22 we'll start in the morning with this. There are three more  
23 subsections yet, and we'll start in the morning. And to get  
24 better conversation, we'll start in the morning on sub-question  
25 E and start with the weight of evidence at that point. Okay.  
26 Everybody will come back fresh in the morning. Okay. So with



1 that, do you need to say anything Fred. Okay. We're adjourned  
2 for today. We will meet a nine o'clock tomorrow morning.

3 (WHEREUPON the meeting was adjourned for the  
4 day)

5 **DR. DANIEL SCHLENK:** As with yesterday, I  
6 think we will skip the introductions and get into our charge  
7 questions.

8 Before we move forward I believe the Agency  
9 had a couple of comments they wanted to make before we move into  
10 the next letter for the charge questions.

11 **DR. THOMAS STEEGER:** Thank you very much Dr.  
12 Schlenk. This is Tom Steeger with Environmental Fate and  
13 Effects Division.

14 Just as a point of clarification in the  
15 discussions as we are talking about higher tier studies and how  
16 they relate to the laboratory-based studies. The intent of  
17 those higher tier studies as described in the White Paper is to  
18 focus in on particular uncertainties that have been identified  
19 in either the lower tier studies or incident reports that we  
20 might have received or open literature studies, so that you are  
21 addressing very specific risk hypotheses and not generalized  
22 questions.

23 Our experience in the agency with other taxa  
24 and certainly our experience with some of the pollinator studies  
25 that have been submitted in the past, is that they are  
26 attempting to answer too many questions and in the process you



1 do not end up with any answers. So please keep in mind that the  
2 intent of higher tier studies is to focus in on particular  
3 hypotheses.

4 **DR. DANIEL SCHLENK:** Dr. Potter, I think you  
5 had a paper you found for your comments yesterday?

6 **DR. THOMAS POTTER:** Yes, I did. This was in  
7 regards to Question 7, and there was a question from the EPA  
8 staff about examining dust emissions and what might be a  
9 reasonable approach to that.

10 I did some digging last night and I found an  
11 article that was published in Environmental Science and  
12 Technology in January of this year, by an Italian research  
13 group. I'll just enter that into the record.

14 It provides some emission factors for some of  
15 the seed coating insecticide, and using the emission factors and  
16 some of the subsequent deposition data that they were able to  
17 obtain we should be able to come up with an exposure estimate  
18 and see how that compares to the value that has already been  
19 used in the White Paper. I'll make sure we get that into the  
20 record.

21 **DR. DANIEL SCHLENK:** Mr. Pistorius.

22 **MR. JENS PISTORIUS:** Thank you. May I add on  
23 this that is also the report of the opponent, whether reported  
24 on a wider scale on issues on the dust and this paper is one of  
25 the issues that is also in this report.

26 And all of this and even more data is also



1 mentioned in this report, which we include in the document.

2 **DR. DANIEL SCHLENK:** Dr. Berenbaum.

3 **DR. MAY BERENBAUM:** Is this the point at which  
4 I can introduce.? No, okay.

5 **DR. DANIEL SCHLENK:** Okay, we left off last  
6 time at Charge Question 13d, so who wants to read in Charge  
7 Question 13e?

8 **MR. KEITH SAPPINGTON:** Please comment on the  
9 proposed use of a weight-of-evidence approach based on  
10 information obtained from multiple tiers of risk assessment for  
11 characterizing pesticide risks to honey bees.

12 **DR. DANIEL SCHLENK:** The lead on that is Dr.  
13 Tarpy.

14 **DR. DAVID TARPY:** Good morning everybody. The  
15 discussants on this particular point just again want to  
16 reiterate that it is always difficult to move from one level of  
17 biological organization to others but we definitely concur and  
18 agree with the weightof-evidence approach by trying to  
19 synthesize the information from these multiple tiers.

20 And so we also thought that high throughput  
21 methods that are reliable and repeatable are obviously going to  
22 be the most valuable and hopefully this discussion from this  
23 week will be able to generate ideas of how to do those types of  
24 screening techniques that account for the biological complexity  
25 that is added by social organisms such as honey bees.

26 But then we also believe that because the



1 basil unit of selection for a social insect like honey bees is  
2 the colony level, we feel that there should be a centralized  
3 intendancy of the weight-of-evidence towards the Tier II level  
4 that is going to be the most informative. This is also going to  
5 be discussed in the next sub-question for Charge Question 13.

6 **DR. MAY BERENBAUM:** Just two additional  
7 comments that we discussed. One is that certain parameters may  
8 be very readily quantifiable and statistically analyzed but they  
9 may not necessarily be as informative as less easily measured  
10 criteria, which is an argument for weight-of-evidence.

11 And in addition the consideration that the  
12 impact that is being assessed is on a workforce rather than  
13 simply an organism, it is not just an up or down, alive or dead  
14 criterion, and it is quality as well.

15 So quality issues are not as readily  
16 quantifiable so weight-of-evidence of multiple indicators would  
17 be particularly appropriate for assessing honey bee pesticide  
18 sensitivity.

19 **DR. JEFF PETTIS:** Just one additional comment  
20 along the same lines.

21 We talked about biological effects and  
22 statistically significant effects and this weight-of-evidence  
23 approach will allow us to consider those biological effects  
24 that, because of sample size in Tier II and Tier III may be  
25 reduced and we cannot reach statistical significance, we should  
26 keep in mind there is biological effect and I think this



1 weight-of-evidence approach allows us to do that.

2 **DR. DANIEL SCHLENK:** Other panel members.  
3 Really, wow. Great.

4 Let me go back to Mr. Sappington, do you have  
5 any questions or clarifications? Okay, great. Let's go ahead  
6 and read in Charge Question 13f.

7 **MR. KEITH SAPPINGTON:** Charge Question 13f;  
8 please comment on how best to characterize overall uncertainty  
9 or weigh different areas of uncertainty in risk  
10 characterization.

11 **DR. DAVID TARPY:** So this again is what I  
12 alluded to in sub-question e that Tier III studies we feel are  
13 much more prone to type II errors and false negatives and  
14 therefore, Tier II studies seem to be much more robust in trying  
15 to reduce uncertainty in this type of risk characterization.  
16 And that is because type II studies are more prone to type I  
17 errors but therefore are much more conservative.

18 Tier I studies, of course, are very robust and  
19 repeatable but at the individual level do not translate as well  
20 to whole colony systems, and so the weight-of-evidence at that  
21 screening level, there ought to be caution in interpretation of  
22 that.

23 So again, it places the real emphasis of  
24 testing and screening at the Tier II level at whole colony in  
25 vivo type settings.

26 **DR. MAY BERENBAUM:** I concur.



1 DR. JEFF PETTIS: I concur.

2 DR. DANIEL SCHLENK: Other panel members. Dr.  
3 Fefferman.

4 DR. NINA FEFFERMAN: Good morning. Just a  
5 brief comment on it; I concur with everything that has been  
6 said. I want to try and disambiguate. In risk assessment,  
7 especially in modeling right there are. I'm sorry.

8 UNKNOWN SPEAKER: Disambiguate?

9 DR. NINA FEFFERMAN: Yes, to choose a part. I  
10 do not actually mean a clarification of something that has  
11 already been said but I mean it using a part of the dual  
12 meanings of the word uncertainty. Did I invent a word?

13 Uncertainty itself has two very meaningful and  
14 very very very functionally different sources. One is  
15 biological stochasticity leading to different ranges of  
16 measurements that are accurate in their measurement and the  
17 other is not knowing what those values would be.

18 And so treating those as one thing can lead  
19 itself to some confusion. I just wanted to make sure that that  
20 was somewhere in the conversation.

21 DR. DANIEL SCHLENK: Other comments?

22 MR. JENS PISTORIUS: When we address  
23 uncertainty, for instance with the investigation of incidence,  
24 that may be used to describe the risk that has occurred under  
25 practical conditions but nevertheless it only has a limited use  
26 for actually the risk assessment.



1           If you cannot prove that because you have the  
2 proper use and you have the improper use. I guess it is the  
3 same, yes like in Europe to assess as a risk assesses the proper  
4 use of the substance. This is basically one uncertainty that  
5 you have to take into account when considering incidents.

6           I think basically you have on all different  
7 tiers uncertainties starting with lower uncertainties from Tier  
8 I; they increase a little bit, Tier II. I think Tier II studies  
9 with worst case exposure have quite a low uncertainty.

10          Depending on the number and the design of the  
11 studies you may have a small uncertainty or medium uncertainty  
12 also in the tier test, if the study design is bad you may have a  
13 very high uncertainty for sure. But it depends on also the  
14 number of studies that are actually handed in.

15          So I think you have to take those into account  
16 but nevertheless even when you have a certain number of  
17 incidents does not have to change the outcome of the risk  
18 assessment necessarily, if it is not proven or if you cannot  
19 link it to a proper use.

20          For instance, we get a small number of  
21 poisoning incidents with a few insecticidal substances, spray  
22 application, every year. But then again in most cases where we  
23 could trace it down - we cannot trace it down in all cases - we  
24 actually found out that it was improper uses.

25          So too high amount applied, wrong crop applied  
26 mitigation measures not used. But on the other hand, especially



1 the incident may be used to give a feedback on how good is  
2 actually the risk assessment to be incorporated in the real life  
3 situation.

4 This is a part which also may give a feedback  
5 on the uncertainty that you create in your risk assessment  
6 process. And then in some cases may even be valid enough to  
7 say, Okay this application is again a risk management issue not  
8 practical for the farmers to achieve.

9 **DR. DANIEL SCHLENK:** Any other comments? Yes,  
10 Dr. Ostiguy.

11 **DR. NANCY OSTIGUY:** I'm not sure if I  
12 understood Jens' last part well enough. And I do not know if  
13 EPA can address improper use, but when you have an overwhelming  
14 - and this of course is a judgment call - a large number of  
15 improper uses, I do think that that has to weigh into whether or  
16 not we are going to use a particular substance.

17 **DR. DANIEL SCHLENK:** Other comments? Mr.  
18 Pistorius.

19 **MR. JENS PISTORIUS:** I think that's basically  
20 then the part of the risk management to see how good it is  
21 actually possible to incorporate this behavior of normal people  
22 into this and it is not enforcement action but again looking  
23 from at all states we have got different responsibilities.

24 And so it is not a risk assessment; it is a  
25 risk management issue and in all cases it is not the federal but  
26 then it is a state issue of ensuring that those enforcements are



1 done; for clarification of my statement.

2 **DR. DANIEL SCHLENK:** Any other comments for  
3 this particular question? Mr. Sappington, any questions or  
4 clarifications? Dr. Steeger.

5 **DR. THOMAS STEEGER:** Just as a point of  
6 clarification on the incident data. We do make an effort to  
7 distinguish where chemicals have been used properly or misused,  
8 and actually a trend in the misuse of a chemical can inform risk  
9 management decisions as well; that can be a problem in and of  
10 itself. Thank you.

11 **DR. DANIEL SCHLENK:** All right, let's go ahead  
12 and move on to our last sub-question for 13.

13 **MR. KEITH SAPPINGTON:** Charge Question 13g,  
14 please comment on how to focus or prioritize uncertainties when  
15 designing and interpreting Tier II and Tier III studies.

16 **DR. DANIEL SCHLENK:** Dr. Tarpy.

17 **DR. DAVID TARPY:** So most of the  
18 recommendations and issues that have been brought up that weigh  
19 in on this particular question, we feel have been discussed  
20 previously so we do not really want to recapitulate them here.

21 But things like ensuring adequate and  
22 sufficient sample size to increase statically power and then  
23 weighing that evidence to focus uncertainties is good.  
24 Replication of study obviously, and focusing at Tier II levels,  
25 at the colony level for testing phenotypic effects, are all  
26 means of really focusing the designs of those types of studies



1 that seems to be a priority.

2 Also, I understand the real need of wanting to  
3 focus on one or two critical measures and hypotheses, but we  
4 have been discussing all week the complexities of different  
5 routes of exposure and different modes of action and different  
6 ill effects, sometimes very subtle on social insect colonies  
7 that can all result in ill health.

8 It is hard to always identify kind of with one  
9 broad stroke that this is the one thing that needs to be  
10 measured and if that is okay then everything else is going to be  
11 okay with honey bee colonies. So I think including those  
12 complexities in these knowing that one size fits all is not  
13 going to be the case in all of these Tier II and Tier III  
14 studies is definitely something to keep in mind.

15 But hopefully, as we will discuss in Charge  
16 Question 14 that there may be ways to try and narrow those down  
17 and identify some of the real key aspects that can be focused  
18 on.

19 **DR. MAY BERENBAUM:** I just want to add one  
20 other factor to address uncertainties in addition to the  
21 geographic replication, season replication. I like us to keep  
22 in mind genetic variability within managed populations of *Apis*  
23 *mellifera*.

24 **DR. JEFF PETTIS:** I concur with what have been  
25 said.

26 **DR. DANIEL SCHLENK:** Other panel members? Dr.



1 Pistorius.

2 **MR. JENS PISTORIUS:** When we focus on how to  
3 prioritize uncertainties when designing and later on  
4 interpreting tier studies, when designing I think that you have  
5 to have a look at the concern that you get from the lower tiers.

6 You have to identify the relevant root of  
7 exposure that is of concern to you. For instance, you get  
8 different toxicity for certain insecticide; you may get  
9 substances with a high or/and low contact toxicity and vice  
10 versa, or substances which have both high levels.

11 Such information from the lower tier basically  
12 is needed and should be written down what the concern is and  
13 then used to be designing and focusing on the design for the  
14 Tier II and Tier III studies. For instance, also what I  
15 mentioned with the sub-lethal behavioral effect, if you have  
16 such a study where you have severe sub-lethal behavioral effect  
17 then you might have to change the design accordingly and maybe  
18 focus differently on the different aspects that you can test  
19 with your Tier II and Tier III studies.

20 What we have not mentioned in this round is  
21 basically, "What are the different options that you can test in  
22 such a Tier III study."

23 We just talked about tier studies and this is  
24 basically standard tier study, but there are additional methods  
25 available and we cannot elaborate on them all in this short  
26 time, which would actually be able to address specific concerns.



1                   We have shortly mentioned, for instance, the  
2                   impact on homing behavior. There are tests available which may  
3                   be used to address such questions.

4                   In a standard field test, according to EPPO,  
5                   for instance, you do a brood assessment according to the leave  
6                   or failure method, you look at all the brood and the number of  
7                   brood and this is basic in estimation. But you can additionally  
8                   combine it with the OECD 75 method where you actually do mapping  
9                   of the brood and follow the individual brood.

10                  So I think this can just be a general comment,  
11                  but I think the level of the concern that you get from the tier  
12                  I study is basically the most important one which help you for  
13                  designing this.

14                  **DR. DANIEL SCHLENK:** Dr. James.

15                  **DR. ROSALIND JAMES:** Jens that was an  
16                  excellent answer I think. My point seems minor in comparison.

17                  I would add to your answer, I think yesterday  
18                  some comments were made to me by the EPA folks that when we had  
19                  discussions about the Tier I experimental design, that we were  
20                  throwing out sort of too many measures that could be taken. And  
21                  that is one way that they could focus down to which of these  
22                  measures are the ones that are most important and base it on the  
23                  results of the Tier I and the Tier II effects that were seen.

24                  The other thing I was going to add about  
25                  helping with designing an uncertainty is proper replication and  
26                  controls. Honey bees are highly variable systems and then when



1 you go to a colony level you really need to make sure that the  
2 experiments are replicated enough and that the controls are good  
3 controls.

4 **DR. JEFF PETTIS:** Just building on those  
5 comments; another good example is if you had a concern raised in  
6 Tier I that maybe there was chronic exposure that was going to  
7 shorten the longevity of adult worker bees and one ramification  
8 for that would be at the colony level maybe overwintering  
9 success.

10 So designing a study to get at where the  
11 colonies could adequately overwinter after exposure, that kind  
12 of thing. And those get longer in duration, they are harder to  
13 control, but they are possible.

14 **DR. GREG HUNT:** Along those same lines, if it  
15 is going to affect individual longevity in Tier II and Tier III,  
16 it is pretty simple to emerge some bees in an incubator, put  
17 those newly emerged bees that are paint-marked in the colony and  
18 then census them over time to get an average longevity.

19 **MR. JENS PISTORIUS:** To add to what Jeff  
20 Pettis has said, I think it's completely right.

21 One idea that might help you in designing, or  
22 looking at when do I have to do an overwintering or long-term  
23 study. This, of course, is basically self explaining, but use  
24 for instance the substance properties; as the persistence and  
25 the degradation and also the mode of application if it is an  
26 acute spray treatment. Also taking into account the information



1 that is available on other organisms or on the efficacy side,  
2 the mode of action that is proposed.

3 **DR. MARTHA SANDY:** I have a question for the  
4 folks here that have the expertise. You are saying the honey bee  
5 colonies are very variable; I wonder how would you design a  
6 study to take that into account? Would you have sister queens  
7 in the different colonies or what would be the ways to do that?

8 **DR. NANCY OSTIGUY:** There are some things you  
9 can do to reduce that issue. You would pick the same line, so  
10 it is all Italians or all carniolans. You would pick sister  
11 queens.

12 As much as possible you would have the  
13 colonies in a similar micro-environment. I'm sure others have  
14 suggestions.

15 **DR. GREG HUNT:** What she said, but you would  
16 have sister queens all mated in the same mating yard. Of  
17 course, they are mating to about a dozen different males so you  
18 have a lot of genetic diversity in the colony but hopefully that  
19 all averages out and then you have of course more replication.

20 **DR. NINA FEFERMAN:** But that all said, you  
21 need the replication of those control samples in order to make  
22 sure that you are not looking just within one line of effect.

23 **DR. STEPHEN KLAINE:** I just wanted to ask, do  
24 we even know enough about the variation in order to do a power  
25 analysis; so we even have an idea of how many replicates we need  
26 in these designs?



1                   **DR. GREG HUNT:** The answer is probably not  
2 because it depends on which study you are doing and some studies  
3 are going to have so much variability due to environmental and  
4 colony dynamics.

5                   I mean, in theory it is possible, but I do not  
6 know.

7                   **DR. STEPHEN KLAIN:** Real quickly, just go to  
8 probably the easiest one or the one that you guys like, the  
9 tunnel study. Do you have enough information about variability  
10 in the tunnel study? You know, how many hives to put in?

11                  **DR. NANCY OSTIGUY:** I am going to disagree  
12 with Dr. Hunt a little bit.

13                  For particular outcome measurements I think we  
14 can do the power analysis just fine. It gets very difficult  
15 with some of the other measures or something that we really have  
16 not been talking a lot about. The things that we have suggested  
17 as possible future ways to analyze what is going on in honey bee  
18 colony, those we do not necessarily have enough information to  
19 do a proper power analysis. But colony survivorship, fecundity,  
20 those sorts of things. There is a tremendous amount of  
21 variability in the data but we can still do a power analysis.

22                  **DR. JEFF PETTIS:** There is a paper out there  
23 Rose et al., I do not remember the year; that was looking at  
24 effects of Bt corn pollen on bees. And they did a power  
25 analysis, I think at the individual bee level, to look at sample  
26 size but also at the colony level, so there is a starting point.



1  
2 And then my other comment was going to be  
3 about the way to standardize colonies. We often will begin a  
4 group of colonies, just a group of 10,000 bees and a queen. We  
5 call it package bees. So they all start with the same starting  
6 population and presumably same disease levels and grow them for  
7 about a two month period.

8 It is always advisable, if you need 40  
9 colonies for the study, start about 50 colonies; because as you  
10 began the study you can go through those and just take the ones  
11 that are of most equal strength. So there are ways to do that.

12  
13 **DR. GREG HUNT:** Unfortunately other sources of  
14 variability will be disease and honey bees have a lot of viruses  
15 with commonly asymptomatic viruses. If I went and tested my 150  
16 colonies now, they all have deformed wing virus. And the ones  
17 that have higher mite levels have bees that show deformed wings.  
18 If I get rid of the mites, they will still have deformed wing  
19 virus but they will look fine.

20 One way to deal with that is to control your  
21 mites and to quantitatively measure your virus titers. I mean  
22 they could have three different viruses in them at one time so  
23 that is a source of variability.

24 **MR. JENS PISTORIUS:** In addition, I think all  
25 has been said, but not by me. That's a saying we have.

26 You made a very good point, Nancy, in saying



1 that for most of the variables there are very good statistical  
2 tests possible with a high certainty.

3 But on the other hand, for instance when you  
4 look at the number of colonies, when you are just looking at the  
5 certainty that you want to achieve, just look at the statistical  
6 numbers you would need, you end up with ridiculous numbers of  
7 colonies that are feasible to be tested. It is not feasible to  
8 test for every test, hundreds of colonies, basically just to get  
9 a statistical significance, but in my opinion this is not  
10 necessary at all to detect biological significance.

11 So I think it has to be looked at with  
12 caution. Where you need really this statistical yes or no and I  
13 think we all agree that we cannot look at colonies just by  
14 evaluating statistics.

15 So I think it is not appropriate to say, okay  
16 for power analysis reasons we need whatever, 40 colonies  
17 preferred because in other circles I know that such numbers have  
18 come up from statisticians who had no experience with bee  
19 trials.

20 So I would like to warn that this may be a  
21 problem when communicating the results. And I think that with a  
22 lot lower numbers preferred you already are able to do a good  
23 test. And probably this is also the same with semi-field test.  
24 You can do statistics, for instance, on mortality, but the most  
25 important thing is that you know a bit of bee biology, that you  
26 have expert judgement and you consider the biological



1 significance.

2 **DR. DAVID TARPY:** I just want to agree with  
3 all of those things but just to kind of step back to the  
4 question a little bit and say how we can focus and prioritize  
5 these uncertainties.

6 I agree that we are never going to reduce them  
7 to real acceptable level without a ridiculously high sample  
8 size; you know all those kinds of things. And so I think what  
9 we need to emphasis from the answers from the previous question  
10 is that we are just going to have to rely on the interpretation  
11 of the given power of a given study and the inherent variability  
12 that is part of that study, that we can try to control by using  
13 sister queens and other means of standardizing, but we are not  
14 going to be able to get rid of all of them. We are not going to  
15 be able to get rid of all of the diseases.

16 And so focusing on statistical power by  
17 increase sample size, replication of study, and trying to  
18 minimize the factors that we can, but looking at the important  
19 biological phenomenon at Tier IIs is really the issue.

20 So I mean we can go on and on about all of  
21 these different things that need to be control, but in a  
22 practical way cannot always. So it is the accumulation of  
23 evidence that is important.

24 **DR. NINA FEFFERMAN:** I normally do not suggest  
25 this especially for practical use because it is not a practical  
26 science, but everything that we have discussed in terms of



1 significance seems to be boiling down to things like power  
2 analysis and p-values, which is a frequent, just statistical  
3 method.

4           There are actually Bayesian methods for  
5 incorporating lack of understanding of variation in the system  
6 that give you, no longer a threshold test for "Yes this is  
7 meaningful; this is not", but an understanding of how meaningful  
8 given the amount of measurement and the amount of the inherent  
9 variation you expect for that measurement, how meaningful the  
10 interpretation of your measure have been.

11           I do not want to go on record as suggesting  
12 that that become part of the standard expectation for these test  
13 but I would like to pipe up and say, "It is not the fact that  
14 statisticians are living in a black box and because they've  
15 never seen a bee, their work doesn't apply."

16           There are methods for this - they are very  
17 good, the times that they have been used. They usually get  
18 used, honestly, at this point in practice only in things like  
19 human medicine or international politics that has nothing to do  
20 with science, kind of sociological studies to lend credence to  
21 some others position.

22           But when these things have been applied,  
23 especially in the areas of risk assessment for then separate  
24 managerial decision making, they have proven to be very  
25 informative. So that may be something to keep in mind maybe in  
26 generations out - it would be a mistake in this conversation to



1 assume that there do not exists techniques to handle this.

2 **DR. DANIEL SCHLENK:** Just to follow up on that  
3 point. I do know having to attend SAP meetings recently that  
4 that is actually being proposed in risk assessment paradigm,  
5 Bayesian techniques actually. That is a huge discussion point.

6 **DR. GREG HUNT:** Yes, Bayesian methods are used  
7 in many areas of biology. But getting back to the power  
8 analysis thing and agreeing with Jeff and Dave that with honey  
9 bee colonies, in Tier III studies, we cannot expect at the  
10 colony level statistical significance. But at Tier II and Tier  
11 III, if you can identify things that are important at the  
12 individual level, you can use statistics at the individual level  
13 to predict.

14 **DR. ROSALIND JAMES:** Do you have a good  
15 general reference on how Bayesian might be used in a risk  
16 analysis, or analyzing this kind of data?

17 **DR. DANIEL SCHLENK:** There will be a report  
18 coming out - well, the draft is being circulated right now, so I  
19 guess in another month, maybe?

20 **DR. ROSALIND JAMES:** Can we put a reference to  
21 that in our notes?

22 **DR. DANIEL SCHLENK:** You could, yeah. Yes.

23 **DR. STEPHEN KLAINE:** There are a number of  
24 publications using Bayesian in terms of uncertainty analysis.  
25 Dwayne Moore and several others have published on this.

26 **DR. ROSALIND JAMES:** That would be relevant



1 for this type of experiment.

2 **DR. STEPHEN KLAINÉ:** I'll get the references.

3  
4 **DR. MAY BERENBAUM:** It is a lit bit alarming  
5 to hear statements that you cannot do statistics on. I just  
6 want to make the observation that there appears to be a  
7 correlation, which is likely statistically between the amount of  
8 media interest in a particular subject and the number of poorly  
9 designed studies that lacks statistical analysis, which gets a  
10 disproportional amount of media attention.

11 So I just want to emphasis that the absence of  
12 a statistical analysis or design that allows statistical  
13 analysis profoundly reduces the utility of any study, as it  
14 should. And this particular field is really prone to  
15 high-profile statistically - for want of a better word -  
16 worthless studies.

17 **DR. JEFF PETTIS:** Just an additional comment  
18 about all of this. We convene Vets to talk about bee diseases  
19 and transmission of other animal diseases, and bees always come  
20 out as the exception. If you are dealing with dairy cow,  
21 chickens, or whatever, they are just so much easier to deal with  
22 because you do not have this individual to colony level effect,  
23 so this is nothing new, and in disease transmission around the  
24 globe and stuff.

25 If you start dealing with any organism, a  
26 flying bird or a land animal, the things are all the same until



1 you get to bees and then all the rules changes, so it is not  
2 unusual.

3 **DR. DANIEL SCHLENK:** Okay, Dr. Tarpy a lot to  
4 integrate into that answer for you so I will leave that to your  
5 best judgment. And again let me just reiterate that if you are  
6 making comments, please get those written comments to Dr. Tarpy  
7 so that he can integrate that into the minutes.

8 Any other panel comments? Okay, let me go to  
9 the agency, Mr. Sappington do you have any questions or  
10 clarifications.

11 **MR. KEITH SAPPINGTON:** No Mr. Chairman we are  
12 good.

13 **DR. DANIEL SCHLENK:** Great. Okay, let's move  
14 on to our last question, 14. You want to read that into the  
15 record.

16 **MR. KEITH SAPPINGTON:** Charge Question 14a;  
17 please comment on the concept of using colony-level ecological  
18 models to inform the proposed risk assessment process for honey  
19 bees, as indicated above.

20 **DR. DANIEL SCHLENK:** And our lead on this  
21 question is Dr. Fefferman.

22 **DR. NINA FEFFERMAN:** So I'm kind of worry that  
23 everyone is going to hate me on this question. My basic answer  
24 to all three parts of this question is, each of these questions  
25 is important but I think you are starting with the wrong  
26 questions. So if you do not mind I am going to start with a



1 sort of general explanation of what I mean by that and then move  
2 into answering the three particulars. Is that okay with the  
3 Chair? Okay.

4 So to start with an idea of methodology and  
5 model choice that we have talked about a little bit earlier this  
6 week of analytic versus simulation models. And it seems that  
7 most of these questions have focus on the stimulation modeling  
8 level.

9 And in my opinion this misses really where the  
10 greatest utility of modeling is going to be for these types of  
11 questions for risk assessment that are being asked throughout  
12 the rest of the White Paper.

13 And that you guys have done an amazing job of  
14 picking out where the simulation type of questions have been  
15 addressed in the literature and where they might be able to help  
16 in the future but that that misses an entire analytic level.  
17 Especially for Tier I studies, I actually think that the need  
18 for simulation modeling might be obviated by the fact that - and  
19 this we will get to later in a sub-question -- by the time you  
20 have measured carefully enough all of the parameters that you  
21 would need and all of the outputs that you would then validate  
22 the model against, you may not need the model to answer the  
23 question anymore. And that for simulation modeling may for Tier  
24 I studies no longer then help you, although for Tier II and Tier  
25 III type equivalents would be very helpful.

26 So to explain analytic models, they are kind



1 of easy examples certainly from physics and I can detail some of  
2 those in the notes, although I probably will not go through them  
3 here. But the example that we discussed yesterday of the ratio  
4 instead of making the direct link from worker mortality for  
5 colony success to the ratio of worker mortality to worker  
6 eclosion rate was a very informative discussion from my end in  
7 terms of what an analytic model can be.

8 That is an analytic model right there saying,  
9 "We need to look at that ratio." Now, of course that ratio is  
10 not all we need to discuss colony health; so for example notice  
11 that one worker dying over one worker eclosing for an entire  
12 colony would keep that ratio fairly healthy looking and that  
13 would be terrible.

14 So we need to have minimum constraints on  
15 things like population sizes; and in some cases we need to have  
16 maximum constraint on population sizes to do things like if we  
17 want to prevent swarming or if we want to worry on a population  
18 level about carrying capacity type issues.

19 But incorporating just those ideas of that  
20 ratio and that constraint without knowing any measurements  
21 already gives us sort of a universally applicable metric of  
22 colony health knowing if the colony is thriving or dying. And  
23 that is the hallmark of an analytically valid model, one that we  
24 can apply to gain insight regardless of the system.

25 And actually Dr. James and I was just talking  
26 a little bit in the hallway afterward and I can put the details



1 of that conversation in the notes, about how this could be  
2 tailored with the inclusion of one other variable to represent  
3 solitary bees as well and their success as a population instead  
4 of as an individual.

5 And that means that we can get from this  
6 somewhat abstract. We are just talking about sort of workforce  
7 and its ability to provide for itself as a sustainable  
8 population. We can get very interesting insights and it is  
9 abstract enough to tell us where the detailed measurements need  
10 to go to inform that.

11 So just off the top of my head right, if we  
12 say, okay there needs to be a minimum colony size -- that is  
13 something we would need to go measure. Is there a minimum size  
14 of the colony under which - and we do not actually need to  
15 measure it, we know that, but you guys see what I mean.

16 So there are ways using these abstractions and  
17 equation-based things that give us thresholds. If that ratio of  
18 worker death to worker eclosion is less than one, the colony is  
19 going to die. There just is not replacement value for colonies.

20 That is a strict threshold, that number is one. Right, if that  
21 ratio is less than one. That does not change regardless of what  
22 the worker eclosion rate is; so before we measure anything we  
23 now know something.

24 So these are a way to get at some of the  
25 conversation we were having yesterday about how we identify the  
26 important things to go measure, where do we go look and the more



1 generally applicability across these things. Of course then you  
2 sort of standardly validate them against expertise to ensure  
3 that you captured the relationships appropriately and they do  
4 still make simulation level predictions.

5 From each analytic model you can then simulate  
6 an outcome. You probably can use an Excel program that captures  
7 that ratio and goes, "Okay, for a colony of this size should it  
8 do okay?" "How long should it take before it is not okay?"  
9 That is a testable hypothesis prediction that leads to more  
10 immediate approximate metrics for how to test, even if those  
11 metrics are not the goal endpoint.

12 So it is actually one of the greatest  
13 utilities of this type of model in addition to providing those  
14 theoretical insights that you then generate incidental things to  
15 measure along the way that do not lead to the outcome of is the  
16 colony dying, but are still testable and measureable.

17 And say, "Okay, if the colony is going to  
18 succeed there should be this other thing that has to happen as a  
19 logical necessity of that successful process. That is not an  
20 outcome we would discuss even normally a biologically relevant  
21 thing, but is a logical equivalent necessity of a good process  
22 happening. Where do we go measure those?

23 This is not to say that this is the equivalent  
24 of a data gap. My saying that the questions and the White Paper  
25 do not address this type of modeling, that is not to say that  
26 there is a data gap here.



1           One of the benefits of this analysis is that  
2   for non-risk assessment, non-pesticide and toxicity purposes, we  
3   have analytic models of healthy honey bee colony processes.  
4   I've written a couple of them and can provide references for  
5   that if that will be helpful, and I will just stick some in when  
6   I write up these notes.

7           Each of those has mechanisms of impact from  
8   different pathways and it would be very easy in a modular sense  
9   to sort of unplug the pathway that is studied in that setting.  
10   Sometimes it is foraging richness and in fields there is  
11   uncertainty there. Sometimes it has been competition, sometimes  
12   it has been predators, any of these sets of things.

13           We can modularly then plug in models that say,  
14   "Well, if the empirical measurement of toxicity says here is the  
15   impact on longevity, here is the impact on colony level  
16   fecundity, not necessary queen fecundity, but colony support of  
17   the eggs the queen lay.

18           If we plug those in modularly to the numerator  
19   and the denominator, here is our outcome for is this colony  
20   thriving. And so there is not necessarily not at all forget  
21   necessarily. There is not a gap in the availability of these  
22   tools; they simply have not been applied directly yet to this  
23   question. So they have not been analyzed to say, "Well then if  
24   I just look at this model, the clear threshold would be blah."

25           But they are easily calculable as soon as  
26   someone turns their mind to it. And as soon as someone has some



1 of the individual and group level measurement that you guys are  
2 discussing getting already, without that necessarily being the  
3 end point. Where we could use those as proximate ways to get at  
4 the ultimate question of what is the success of a colony and  
5 what is the success of a population at providing pollinator  
6 services, at being sustainable itself and having population  
7 level success.

8 Lastly, in the general sense - sorry I realize  
9 this is a very long answer -- in the general sense modeling does  
10 create one set of complications that nothing else we have  
11 discussed creates for the levels of the Tiers. It is important  
12 to point out that the tiers themselves are somewhat problematic  
13 in the sense that the kind of analytic modeling that I am  
14 suggesting would be useful.

15 Because colony level models should ideally be  
16 used backwards to inform which Tier I types of studies should  
17 happen to be most useful as a pass; by which if you pass Tier I  
18 and you do not set off any alarms to trigger further study,  
19 those were your important metrics. Those need -- not semi-field  
20 and field, in that sense I do not mean tier, but because of the  
21 way that these study works we have been discussing Tier II and  
22 III models as being the ones that leads to colony level and  
23 population level type questions, because those are hard to get  
24 to in a lab.

25 So that equivalence of which tiers mean which  
26 level of scale, no longer applies within the modeling. And a



1 Tier I study should possibly be, even a population of colonies  
2 level model in order to backwards inform what the individual  
3 honey bee or solitary bee metrics should be that are most  
4 critical to indicting health, and I mean health in a  
5 sustainability kind of sense.

6 Very briefly then to address the specific  
7 question of (a) which was the concept of colony-level ecological  
8 models, yes it is great. The short answer is we should do it.  
9 We should do it in intelligent ways. So exactly the types of  
10 questions that we have been saying are very hard to get to  
11 experimentally of how do individuals scale to colony, scale to  
12 populations, and how does all of that go as things fluctuates  
13 over time both in terms of natural stochastic fluctuations and  
14 in terms of bees changes.

15 The year changes, bees changes. The need  
16 changes; bees have different behaviors if a bear comes by, that  
17 kind of stuff; although agricultural bears are probably not a  
18 big problem, but still.

19 So these kinds of models are simulation and  
20 analytic but especially simulation, the kinds of models you  
21 proposed directly in the paper, are going to be quite useful for  
22 doing that. All this being true, there is a huge pitfall to be  
23 avoided in doing this. It would be a mistake to just suppose  
24 that the best modeling practice for colony-level ecological  
25 models to inform risk assessment would be an all-inclusive, it  
26 has every aspect of colony function and every risk model. That



1 is a bad way to model this even in simulation.

2 And that has to do both with being able to  
3 interpret the outcomes and knowing which factors were driving  
4 them but also as we just addressed in some of question 13, there  
5 is a confounding uncertainty from each modular plug-in. And  
6 that is both the uncertainty of the measurement and the  
7 uncertainty of the stochasticity.

8 And there are really intricate ways in which  
9 those interact and there are some tools from statistical  
10 mechanics to try and correct for that, that we do not want to  
11 use for this.

12 And so a much better way of approaching an  
13 all-inclusive type of model is to have littler models as some of  
14 are in the White Paper discussed, and then have a risk assessor  
15 as a person with expertise decide how the outcomes from those  
16 really might interact. Sticking them all into one model where  
17 the math comes out as one number is a great way to confuse  
18 ourselves.

19 That is basically my answer to a.

20 **DR. ROSALIND JAMES:** So my role on this team  
21 was to help Nina give examples where things were not clear. So  
22 I am going to ask you a couple of questions that I had.

23 When you are talking about the tier test you  
24 said it slightly backward. I wanted to make sure you understand  
25 in the tier testing things move to higher tiers when they fail  
26 the lower tier, right?



1                   **DR. NINA FEFFERMAN:** Sorry, I misspoke. What  
2 I mean is if there is a trigger that says I failed Tier I.

3                   **DR. ROSALIND JAMES:** So then a higher risk,  
4 right, not less risk?

5                   **DR. NINA FEFFERMAN:** Yes, right.

6                   **DR. ROSALIND JAMES:** Okay, I wanted to make  
7 sure that you understood that.

8                   And so I was not exactly sure what you were  
9 saying regarding the tier-level and maybe to help clarify I will  
10 put it in another question. Could you go back and look at  
11 current colony-level data that we already have for pesticides  
12 that are in the literature to go back and help determine what  
13 Tier I factors we should note?

14                   **DR. NINA FEFFERMAN:** Thank you Dr. James so  
15 much. Yes, that is exactly what I meant we should be doing.  
16 That we should take the colony-level understanding that we have  
17 and any future colony-level understanding from whichever either  
18 Tier II or Tier III studies are proposed, if things triggers  
19 higher concerns from Tier I; to go back to suggest further  
20 refinements in which Tier I should trigger further concern.

21                   Does that help clarify?

22                   **DR. ROSALIND JAMES:** I think so, but could you  
23 use an example?

24                   **DR. NINA FEFFERMAN:** For example, and this is  
25 a silly example because it will wind up seeming obvious but  
26 there are others. Actually, can I depart from honey bees



1 comfortably for an example?

2 Okay, awesome. So we have been talking about  
3 some desert tortoise conservation issues and doing some of the  
4 desert tortoise conservation work - I was helping model some of  
5 this -- we went initially to a bunch of desert tortoise experts  
6 and said what do you think are the most salient features of the  
7 endangered population of desert tortoises we are working with.  
8 And they gave us a list of here are the ones we think are really  
9 important and then here are the ones that we think are sort of  
10 ancillary but tortoises depend upon these things.

11 By looking at the population level  
12 determinants of these factors we were able actually to move some  
13 things out of the categories that the experts expected were  
14 going to be very important. Saying, okay, yes that is clearly  
15 killing tortoises; that is not the thing driving population  
16 decline. That is some of the things you are seeing when you go  
17 find a dead tortoise; that is what you are seeing the dead  
18 tortoise do. But it turns out that the value of that age-class  
19 to the demographic process of population growth, it is not  
20 valid.

21 On the other hand, there were some things that  
22 were unexpectedly incredibly important. Water reservoir  
23 balance, if you have a respirator infection as a tortoise it is  
24 apparently really important for conservation. And water balance  
25 was known to be important. Respiratory infections were not  
26 necessarily. And the fact that you get a runny nose turns out



1 to drastically change your water balance.

2 Fugitive dust was assumed to be a very  
3 important thing. Fugitive dust, you have an ATV kick up dust in  
4 the desert and then it doesn't settle for a while, turns out  
5 basically unimportant for our population.

6 So is that a kind of concrete example where,  
7 from going from the population level in modeling, what is the  
8 demographic growth process of that population.

9 We were then able to go back to these  
10 individual measurable things either about the environment or  
11 about the individual tortoise. And say okay, these are the  
12 things you should measure. If you want to be concerned about  
13 your population or if you want to be confident your population  
14 is okay.

15 Does that clarify?

16 **DR. ROSALIND JAMES:** So here is the case where  
17 you could go back. Now the next endangered species. What did  
18 you learn from that that you can say now for Tier I test we want  
19 to measure?

20 **DR. NINA FEFERMAN:** So the process of that  
21 modeling is the thing that ports across species. But it would  
22 be true that we would reformulate the models themselves if we  
23 went from - I mean, if we look at something that is still like a  
24 tortoise then it would still apply. Sure, for other tortoises  
25 not the desert tortoise.

26 Then we could re-apply the same logic and use



1 the same parameters and that does give us thresholds for which  
2 measurements. So we could say, well when we stick in the value  
3 for -- here is for example the non-stable age demography. It  
4 turns out that many of the models -- the outcome is very  
5 sensitive to how many juvenile you have versus how many  
6 reproductive adults. And it turns out that the size of an adult  
7 female is a really important thing for tortoises because eggs  
8 have to fit under the shell.

9 But that turns out to be important in the  
10 demography we had for our population. We would reapply that  
11 analysis in a new population and say well how many adult  
12 tortoises do you have? If you have zillions of adult female  
13 tortoises, then female tortoises' mortality might not be the  
14 issue; it might be egg survival. And that is putting in new  
15 number to the same analysis to get out a different insight for  
16 that population, or for that species.

17 That model would not directly apply to  
18 colonies of honey bees but the exact same methods would and we  
19 do have colony and population-level models of honey bees that do  
20 equivalent things. It is just that since I have not done them  
21 in this context of toxicology I do not have as ready yet, an  
22 answer for like, "this is a surprising result I expect to get  
23 out" because if I expected it, it would not be surprising.

24 **DR. DANIEL SCHLENK:** Okay, let's go to Dr.  
25 Potter.

26 **DR. THOMAS POTTER:** Well I certainly agree



1 with my colleagues and concur that it is important to have a  
2 multi-dimensional approach to the problem, if I am correct in  
3 paraphrasing here. And certainly there are many different types  
4 of models that can be used, again both analytic and simulation.

5  
6 To further try to compartmentalize this  
7 problem, which obviously is exceedingly complex, we can -- again  
8 as Nina mention there are modular approaches that can be used  
9 and at minimum I believe we need to step back and say you know  
10 this is a two-part problem.

11 And forgive me for stating the obvious, but it  
12 is exposure and response. So I would say that as a first cut we  
13 divide our problem down into two parts, number one exposure and  
14 response.

15 Exposure is probably more amendable to  
16 simulation modeling I would hazard a guess, maybe, maybe not.  
17 It certainly has a long history in all of the other aspects that  
18 the Office of Pesticide Programs addresses in terms of  
19 evaluating exposure; certainly simulation models are critical  
20 tool, certainly on the exposure side.

21 And so I strongly would emphasize that effort  
22 be directed in that area to develop simulation models, whether  
23 they be pesticide Environmental Fate model that works for a  
24 colony in terms of residues that are in the colony and  
25 determining what their fate may be in terms of persistence,  
26 longevity, whatnot. Obviously those are critical questions that



1 need to be answered in terms of exposure at different  
2 life-stages, et cetera, overwintering success.

3 And then the other part of that problem, and I  
4 think again Dr. Fefferman identified this and I think it is  
5 critically important and an exceptionally useful tool, perhaps  
6 at Tier I, and that would be the use of inverse modeling to try  
7 to do some ground-truthing, particularly in the side of the  
8 exposure pathway.

9 Again, obviously as has been discussed over  
10 the last few days, we have some uncertainty about exposures.  
11 Efforts have been made to identify where those are. One  
12 possible approach to get at this problem is to take existing  
13 colony data and do adverse modeling; looking at the different  
14 compartments where residues are stored within the colony and  
15 inverse model to see if there are insights to be gained on where  
16 those residues may have come from, whether they are potentially  
17 dust emission.

18 Again, there are some timing issues and  
19 whatnot here et cetera, whether they came in by water or they  
20 are totally dietary. I think to some degree we can gain insight  
21 by going through that process.

22 I will just sum up that that is obviously a  
23 big problem. We recognize that and we would benefit  
24 substantially by subdividing it and proceeding forward from  
25 there.

26 **DR. DAVID TARPY:** I agree with and very much



1 appreciate this discussion and really have nothing more to add  
2 other than to emphasize the previous comments that were made  
3 that colony-level modeling is very helpful in a kind of a  
4 top-down approach. I think the statement was made earlier that  
5 predictive modeling from lower tiers to upper tiers is something  
6 that we need to be very cautious about.

7 And so just simply taking information from  
8 sub-colony phenomena and trying to model that upwards to colony  
9 and population-level is froth with potential problems; rather  
10 taking a colony-level top-down approach of trying to understand  
11 that phenomena that informs the lower tiers is going to be much  
12 more effective.

13 **DR. DANIEL SCHLENK:** Other panel members.

14 **MR. JENS PISTORIUS:** 1:02:08 I wish I had only  
15 a fraction of understanding of modeling of what you guy do. I'm  
16 really impressed and my brain is working a lot at the moment and  
17 I might be confused so excuse me in advance if this answer may  
18 sound funny and some things may have been said. My answer will  
19 be short.

20 Yes, to inform and to identify potential risk,  
21 which trigger maybe the need for higher tier tests; but I want  
22 to point out that, no for if you have a model that tells you if  
23 you identify a risk in Tier I, and then you come up with a model  
24 calculation that said well you do not need a Tier II and Tier  
25 III, I think this would be a funny way you would have two  
26 complimentary ways in risk assessment, which somehow does not



1 fit in a risk assessment scheme.

2 **DR. NINA FEFFERMAN:** Mr. Pistorius, I am  
3 probably misunderstanding. I think that what you just said is  
4 that you should only use modeling for what to do in Tier I if it  
5 agrees with the empirical work. That cannot be what you meant.

6 **MR. JENS PISTORIUS:** I'm not quite sure if I  
7 understood because again my brain might be overworked at the  
8 moment.

9 I just want to say, if you identify a risk in  
10 the known Tier I studies and if you have this risk assessment  
11 process as it is outlined now and if you trigger higher tier  
12 tests. And then if you come up in parallel with the modeling  
13 that says, okay but all you have done in your Tier I  
14 identification of a potential risk is all not valid. And then  
15 you basically have to decide okay our Tier I process identified  
16 would have to go to a higher tier study.

17 And then on the other hand you come up with a  
18 model that says, well yes you have identify the risk in Tier I  
19 but actually it is not necessary, you do not need to do any  
20 further studies. That is something I would reject because then  
21 you would have two parallel ways in risk assessment.

22 Was that clear and understandable?

23 **DR. NINA FEFFERMAN:** I think I understand you  
24 but disagree with you. I think that in my mind that is exactly  
25 the use of half of one of the ways that this works.

26 One would be to identify Tier I things that



1 should be measured empirically that would then trigger the need  
2 for higher studies by concern; but the other would be to show  
3 when we think things that need to be measured that would trigger  
4 higher concern should actually not effect success.

5 And if we reject that half of the modeling  
6 then I do think that we disagree. I think that is still a use  
7 for modeling.

8 **DR. MAY BERENBAUM:** Good luck explaining that  
9 to the public. That is an issue that exactly addresses your  
10 point that if there is a cause for concern generated by  
11 empirical data at Tier I and a decision is made based on models  
12 not to proceed, at lease for the American public, that would I  
13 think create a major concern.

14 **MR. JENS PISTORIUS:** I think it is the same  
15 for the European public but also for me as a person  
16 understanding. Sorry that I really have to say I disagree.

17 I want to see studies with bee colonies and  
18 because we have identified that bee colonies are a lot more  
19 complex than the individual and modeling may be very good and  
20 may inform a little bit, but if this decision is to be made and  
21 say, okay you have identified a risk with a normal risk  
22 assessment Tier I process, if model comes up to say, well you do  
23 not have to do higher tier studies, I would clearly reject that.

24 **DR. NANCY OSTIGUY:** I think that this  
25 discussion points out the importance of doing the analytical  
26 models first. And we really should do those first so we can



1 then target which Tier I studies are the most important to do,  
2 so we do not have contradictory conclusions that we have to  
3 resolve.

4 **DR. MARTHA SANDY:** Just to remind everyone,  
5 risk assessment is -- if you remember, the framework diagram we  
6 saw - it is supposed to be an iterative process. And I think  
7 that that is where we have some confusion going on.

8 I think what Nina is suggesting is, we have  
9 come up with some Tier I endpoints we think are important to the  
10 colony. She is suggesting we use the analytical models to see  
11 if those endpoints in fact are relevant to colony health. And  
12 she gave the example using a different species.

13 But to take another example from the honey  
14 bee, what if we think mortality to drones is an important thing  
15 but in the model it turns out that that does not seem to be a  
16 driver to colony health but what does seem to be a driver is  
17 mortality to larvae and length of time of development of larvae  
18 for example; those are really key.

19 Then what you would do is you would take that  
20 information from the model, once you felt confident that you  
21 could rely on it, you might go back and revise the Tier I test  
22 requirements with which things in Tier I endpoints the  
23 measurements that you place and emphasis on that triggered going  
24 to the Tier II or Tier III test.

25 So I think you are talking at cross-purposes  
26 because you are not, right now Mr. Pistorius, ready to accept



1 that the modeling has been validated and everyone has agreed to  
2 change the current Tier I endpoints that are measured.

3 We are not there yet; we have not even done  
4 the modeling. But, there would be a period of time of  
5 discussion and then different groups would decide whether they  
6 still want to evaluate a certain measurement.

7 **MR. JENS PISTORIUS:** Well I think such  
8 modeling are beautiful for the weight-of-evidence and the  
9 multiple information that you need for the conclusion on the  
10 risk assessment; on the other hand from my experience it is not  
11 that an applicant comes with one study and then you get concern.

12 On the Tier I level, usually what we get is a package of the  
13 standard requirement, which are in our case oral contact and  
14 maybe in future the larvae if we are able -- or larvae will be  
15 addressed as we get there.

16 So that is basically the standard package we  
17 ask for to be able to make a first conclusion. And then, okay I  
18 admit maybe such models may inform do you need other studies on  
19 the tier level one risk assessment. But still on the first  
20 three that you get, on the contact, oral and larvae, you already  
21 do make a decision, you know, are we having a concern?

22 And so from my understanding, but I may be  
23 completely wrong, that is already triggering something. And I  
24 am really glad if you say as a risk assessor, okay we have  
25 identify risk in Tier I, we go to Tier II and we ask for  
26 semi-tier study and we find no effect.



1                   And then you say okay for our  
2 weight-of-evidence you take those multiple information into  
3 account and then you can say with addressing the uncertainties  
4 that you get in your risk assessment and say okay with this  
5 weight-of-evidence we have the Tier I, Tier II and then we find  
6 no effect and this is basically also what the colony said, so I  
7 think you have a lot of weight.

8                   That is my simple understanding of the issue.

9                   **DR. NINA FEFERMAN:** Thank you very much Dr.  
10 Sandy. I think the iterative process is what I have in mind and  
11 it sounds to me like what you are suggesting is that you would  
12 trust modeling to suggest additional test but not to eliminate  
13 existing ones. And I think then we just have a difference of  
14 opinion on the utility of models.

15                   But to address Dr. Berenbaum's comment about  
16 could the public ever accept this, the answer in my mind is  
17 salesmanship. We have a couple of examples where we botched it  
18 entirely and a couple of examples where we have done it really  
19 well.

20                   We botched it entirely with vaccine safety; we  
21 have also botched that with empirical studies. So just having  
22 empirical studies is also not a guarantee of public acceptance  
23 of the results.

24                   But we have done it really really well with  
25 some of the U.S. Fish and Wildlife Services Conservation  
26 Endangered Species Act requirements. And when you should close



1 roads that inconvenience people in order to left spotted  
2 salamanders cross and things like that. And we have had really  
3 good public acceptance and that spotted salamander work, all  
4 modeling, all of it.

5 And it originally met with huge public  
6 resistance and we did really good outreach. And we went in and  
7 we said, okay guys this is what went into this. We did not  
8 explain the equations but we explained the conceptual flow of  
9 what we were modeling and we now have communities that come back  
10 to us every year and go, "Should we be closing the same roads at  
11 the same time this year?"

12 And so my answer to that part is, that is a  
13 matter of public outreach and effective communication. It is a  
14 slightly harder sell to sell the public on modeling versus  
15 empirical work, but forgive me, I have great faith in the  
16 public; I do not have great faith in the average person as a  
17 scientist.

18 So I think we have work to do to explain  
19 empirical science to the public, also we just have a little bit  
20 more work to do to explain mathematical science.

21 **DR. THOMAS POTTER:** I just want to make a  
22 comment. I'm not sure I heard anywhere in the discussion,  
23 certainly in Dr. Fefferman's answer at the outset as well as the  
24 other discussants, that we as a group or anyone was recommending  
25 that modeling be used to in some way subvert or eliminate or in  
26 otherwise change the tier risk assessment process.



1                   We have Tier I, if you fail Tier I one assumes  
2 then you get to Tier II. And certainly, this again, there is a  
3 long history of that in terms of looking at other problems that  
4 the agency deals with. I appreciate your concerns that somehow  
5 an ill-informed model may somehow obscure the need for further  
6 testing but I do not think that that was in any way suggested.

7                   **DR. JAMES MCMANAMAN:** So I got really  
8 confused. I really appreciate your discussion Nina.

9                   It appears to me that (a) is asking a  
10 different kind of question than what we have really been  
11 discussing. Because if we take your examples of analytical  
12 modeling - and I may be way off base here -- if you take your  
13 examples of analytical modeling and apply them to what are the  
14 causes of loss of bees, I think that the European data would  
15 suggest that pesticides are a cause of loss of bees.

16                   If the question is then how should we model  
17 pesticide effects on bee colonies, then it seems to me it is a  
18 different kind of modeling process all together. Is that not  
19 the case? Because in one case you are asking, is it a risk  
20 factor, and if the answer is no, then we have eliminated the  
21 entire question. So if the answer is no, but we still want to  
22 know more about how pesticides affects bees, then I think it  
23 seems to be more of a different type of modeling rather than  
24 analytical type.

25                   **DR. NINA FEFFERMAN:** Thank you. That is a  
26 wonderful clarification question.



1                   When I was talking before about colonies  
2 thriving or dying as an outcome, I should have been more  
3 explicit. I meant that restricted to the example I was using  
4 from the discussion from yesterday, of what is the importance of  
5 a particular threshold level in worker mortality.

6                   But I do mean to suggest that analytic models  
7 themselves are going to directly and validly, in risk  
8 assessment, be able to point to us the most important either  
9 that variables or functional relationships or thresholds for  
10 where pesticides can affect success by whichever metric we  
11 adopt, either pollinator services or survival of the colony or  
12 sustainability of the population.

13                  At any level of those analytic models will be  
14 able to suggest to us -- for it to be a good model both testable  
15 predictable outcomes that should be before we get to the outcome  
16 that we are interested in so that we can validate the model and  
17 not necessarily on an agreed outcome of what we want but  
18 proximate measures that should be easy to measure also easy to  
19 see when the answer is negative. They should not be patently  
20 obviously always going to be true.

21                  So a way of validating, if you would, the  
22 by-products of the model as a way to have confidence in the  
23 entirety of the model when we are asking these difficult  
24 questions; but then apply it to gain insight into which things  
25 should be measured that will directly affect whether or not a  
26 pesticide is posing a risk to our outcome. Thank you so much.



1                   **DR. JAMES MCMANAMAN:** Thanks for the  
2 clarification.

3                   **DR. DANIEL SCHLENK:** Okay, any other comments  
4 right now? I'm going to try to bring us back hopefully. It  
5 seems like all three of the questions have sort have been  
6 discussed at the same time interactively here. So let's go back  
7 to (a) and just say, please comment on the concept of using  
8 colony-level ecological models to perform proposed risk  
9 assessment for honey bees.

10                   Okay so we have had comments, and it appears  
11 that in general the panel feels, yes definitely use models. I  
12 think there is some disagreement perhaps in what outcomes those  
13 models could be used for, perhaps, so let's make sure we get  
14 that into the minutes in terms of concerns for certain panel  
15 members and others.

16                   But it would appear that everybody is  
17 comfortable with the concept of using models for this particular  
18 parameter. Is that what I'm hearing? Yes, okay.

19                   All right, any other comments before we ask, I  
20 am sure there is going to be questions of clarification from the  
21 agency, I am guessing. Any other comments before we go to the  
22 agency?

23                   **DR. THOMAS STEEGER:** I would just like to make  
24 a comment and I appreciate the dialog that is going on regarding  
25 how you interpret information across multiple levels of  
26 biological organization.



1                   The National Research Council through the  
2 National Academy of Science in their publication, Toxicity  
3 Testing in the 21st Century, has sent a strong message that  
4 there should be movement away from whole animal testing and  
5 taking advantage of as much of the available information as you  
6 can so that you can move from one level of biological  
7 organization to another without as much reliance as we have in  
8 the past on whole animal testing.

9                   And that particularly deals with the screening  
10 level approaches where that would enable of regulatory authority  
11 to determine where you need to best use resources and apply  
12 them.

13                  And this whole idea that we touch on in the  
14 paper, adverse outcome pathways, expert systems, that once those  
15 relationships become clear and you can, based on the mode of  
16 action of a chemical, you can have a mechanistic approach that  
17 would enable you to make some decision that would be less  
18 reliant on animal testing, is the direction that this agency I  
19 believe is headed.

20                  But with that said, where you have identified  
21 risks you would be transitioning likely to higher tiered test to  
22 substantiate these potential concerns. Because companies do put  
23 a lot of money into the development of chemicals and it is  
24 ultimately up to the risk manager to decide where a higher tier  
25 testing is required but it is also the purview of the regulated  
26 community to demonstrate their product is safe as it is actually



1 planned to be used. So the higher tier testing would still have  
2 a role.

3 But I applaud the efforts to introduce this  
4 idea of using information at multiple levels of biological  
5 organization to inform the risk management decision.

6 **DR. DANIEL SCHLENK:** Okay, so with that I am  
7 assuming you got what you need for this particular letter. Is  
8 that correct? Okay. With that let's just take a break I think  
9 before we go on to (b) and (c).

10 (Brief recess.)

11 **DR. DANIEL SCHLENK:** Okay, just to remind the  
12 panel, during breaks if you do have any interactions with the  
13 audience or the agency just to make sure that we state that for  
14 the record that you have had those interactions and if it has  
15 changed your comments, you need to state that for the record as  
16 well. Just to let you guys know we could do that.

17 **DR. MAY BERENBAUM:** Do you want all  
18 interactions recorded or only those that have any bearing on the  
19 discussion.

20 **DR. DANIEL SCHLENK:** Those that affect your  
21 comments please.

22 **DR. ROSALIND JAMES:** Do you want those now?

23 **DR. DANIEL SCHLENK:** I will give you the  
24 opportunity as soon as we get to the questions, yes. Let's go  
25 ahead and read letter (b) into the record.

26 **MR. KEITH SAPPINGTON:** Dr. Schlenk, if I could



1 ask one clarifying question.

2 I think it would be helpful to the agencies in  
3 responding - and I'm not sure which of these questions or all of  
4 them - about the difference between an analytical model and a  
5 simulation model and perhaps a concrete example, and perhaps  
6 when one may be preferred over another.

7 **DR. DANIEL SCHLENK:** Dr. Fefferman.

8 **DR. NINA FEFFERMAN:** Thank you for asking that  
9 question and actually we did have a useful conversation on  
10 exactly that point during the break. So I will bring up all of  
11 those points but this is my active disclosure on that one. Do  
12 you want me to answer or just put it in the minutes?

13 **DR. DANIEL SCHLENK:** No, I think just the fact  
14 that you can do that would be appropriate.

15 **DR. NINA FEFFERMAN:** Oh, okay. Yes, we can do  
16 that.

17 **DR. DANIEL SCHLENK:** Okay, ready to go on to  
18 letter (b)?

19 **MR. KEITH SAPPINGTON:** Question 14, letter  
20 (b); please comment on the state of the science regarding  
21 available honey bee models discussed in Section 5.4.2 of the  
22 White Paper in relation to the potential application in a  
23 regulatory risk assessment context. In particular, please  
24 comment on the extent that such models have been evaluated using  
25 empirical data related to honey bee population dynamics and the  
26 availability of such data for their parameterization.



1                   **DR. NINA FEFERMAN:** I going to rely on the  
2 answer to the last question, which is going to come down to for  
3 analytic models data is less important than for simulation  
4 models. And with that context in mind many of the models  
5 mentioned in the White Paper have only very sparse assess to  
6 data thus far. And we have discussed during this panel many  
7 good ways to try and fill some of those data gaps.

8                   And while it is very important to collect that  
9 further input data, for measurements, for parameter values, and  
10 for model validation and to characterize functional  
11 relationships. Many of these analytic models are useful in the  
12 absence of direct measure data as long as the functional  
13 relationships themselves are hypothesize correctly by the  
14 experts building them.

15                   And so this does not happens in a vacuum of  
16 data but what happens is that there is empirical work that  
17 informs relational data. So how do we get - on a very stupid  
18 level - how do we get a bigger colony? We have more babies than  
19 we have deaths, right? That is something that at some point  
20 someone measured and went, "Oh look, demography, demography  
21 happens" when there are more births than death. From that you  
22 get an abstract analytic model, okay births has to outweigh  
23 deaths for a population to grow; the equivalent for that has to  
24 happen for a colony for worker mortality to worker eclosion.

25                   That is a valuable insight; that one is a  
26 trivial insight but valuable insights come out of that



1 relationship in the absence of, what is the worker mortality  
2 today; or what is the overall colony survival with that.

3 So the conclusion of these models to provide  
4 insights into how the systems work and to make specific  
5 predictions about which focal metrics are important, are really  
6 the utility of the analytic side for purposes of risk  
7 assessment. The more general employment of models that can help  
8 assess the importance of unknown levels of threat involves both  
9 the analytic and the simulation levels of these things.

10 So even if very little insight is known about  
11 individual sensitivity to -- for instance exposure to particular  
12 chemicals what you can do is know which pathways to effect,  
13 either within the analytic models that are more abstract or the  
14 empirical models that are the specific. Okay, the link is  
15 multiply this number by .003, get that number that is the  
16 answer.

17 Within the narrow context of the empirical  
18 models discussed in the White Paper, the simulation type models  
19 -- in modeling, simulation models are also called empirical  
20 models, which is not to be confused with empirical experiments.  
21 Sorry, I am going to try and remember not to call them that.

22 Simulation experiments, in the narrow context  
23 of the simulation models being used we then need to discuss the  
24 appropriate sensitivity of the data that is being used, the  
25 sensitivity and robustness testing for the individual  
26 parameters. So there is sensitivity which is how do the value



1 affects the ultimate outcome and there is robustness, which is  
2 what are the range of values for which this model is still  
3 valid.

4 And for a simulation model we have not  
5 discussed robustness at all. There are very good local range  
6 simulation models that tell you exactly as long as PH does not  
7 go off the scale everything in the system behaves exactly this  
8 way; but as soon as you have a PH under three, oh my God do not  
9 use this model; use a different model.

10 There is a different behavior, in physics it  
11 is called a phase transition, right, something else has  
12 happened. You have coalescence or rain from a cloud, right.  
13 There is water, suddenly something happens and water falls out  
14 of the cloud. A model for how water is suspended in the air  
15 works really well up until a point, and then you need a model  
16 that says okay how does rain falls. And I am not a  
17 meteorologist so they might actually have a good model that does  
18 both of those; I do not know.

19 So within the available data that we have  
20 first we have to ask sort of what do we want out of the model;  
21 because if the answer is most appropriately answered by an  
22 analytic model, we may not need any of the data; what we want is  
23 the insight and that will tell us which data then is going to be  
24 important, and then we know whether or not we have it. But we  
25 do not know whether or not we have it until we have that  
26 analytic model to tell us.



1                   For a simulation model we do have to go  
2 through the sensitivity of each of the values that we would need  
3 and say, well what if this was everywhere from here to here.  
4 Does that change the outcome at all? If the answer that comes  
5 out of the entire model of the simulation does not change with  
6 regard to the quality of the data input for that parameter, you  
7 do not need a better measurement of that parameter. On the  
8 other hand if it is very sensitive to that parameter, you need a  
9 very good set of empirical experiments to do that.

10                   So unfortunately there is not an easy answer  
11 to, oh for all of the ones in the White Paper here is the data  
12 gap, here is what we know; we are good just use those. Go  
13 measure those and at the end we will be fine. On the other hand  
14 there is sort of an algorithmic approach that we can take with  
15 each parameter for each model going, is it sufficient; is our  
16 knowledge good enough to use that with confidence in this  
17 context.

18                   **DR. ROSALIND JAMES:** I'm debating whether just  
19 to say I concur or try to elaborate more. I'll try to give a  
20 little example maybe.

21                   In the White Paper I think the review of  
22 simulation model is excellent and fairly complete; there may be  
23 some missing models but I think it is a fairly complete review  
24 and a nice summary of the model.

25                   An example I think of what Nina is talking  
26 about is say with the Khoury et al model, we discussed this



1 earlier and one of the conclusions was there is this critical  
2 threshold on colony of .355 bees per day. If you lose a third  
3 of the bees per day then the colony will collapse but that may  
4 actually be partially dependent on what the reproductive rate of  
5 the queen is in that colony.

6 So that would be an example of a type of  
7 relationships we are talking about.

8 **DR. THOMAS POTTER:** In direct response to the  
9 question I'll say that I feel in the case of exposure modeling  
10 and for pesticides the state of the science is not well  
11 advanced.

12 **DR. DAVID TARPY:** I have no additional  
13 comments.

14 **DR. DANIEL SCHLENK:** Other panel members.

15 **MR. JENS PISTORIUS:** I also have this feeling  
16 that they are valuable to be considered but that they are not  
17 well enough advanced to really be used in the risk assessment.

18 They may inform the risk assessment very well  
19 but for the available models I think all have still some lacks  
20 which really make potentially use very valuable for this moment.

21 Especially the valuation I think is missing and I think that it  
22 has been said for some questions before that the honey bee  
23 colonies, they are a very complex organisms and there are so  
24 many different factors that I think this will always limit the  
25 possibility to describe everything that happens with the bee  
26 colony.



1                   Nevertheless, I think there is some use but I  
2 also know there have been recent publications and also for the  
3 query model in recent discussions we did identified that there  
4 is one important factor missing but at the moment I cannot  
5 recall which one it was exactly, but I can write that into the  
6 notes too.

7                   So I think at the moment the models that we  
8 have are useable to inform risk assessment but again like with  
9 the comment on the question before, I think the most important  
10 is that we still rely on the higher tier test.

11                  **DR. DANIEL SCHLENK:** Other comment. No one  
12 else? Okay. Mr. Sappington do you have any questions of  
13 clarification?

14                  **MR. KEITH SAPPINGTON:** I think we are good,  
15 thank you.

16                  **DR. DANIEL SCHLENK:** All right, we are on our  
17 last question, I think, perhaps, maybe. You want to read (c)  
18 into the record please?

19                  **MR. KEITH SAPPINGTON:** Question 14, final  
20 subpart; please comment on the important elements that should be  
21 considered in reviewing available honey bee colonies, ecological  
22 models for potential application in risk assessments.

23                  **DR. NINA FEFFERMAN:** The first two questions  
24 that should be asked about any model are what insight does it  
25 provide that accurate empirical studies could not, or if there  
26 are none, how does the model spare efforts or expense to provide



1 those insight?

2 In the case of risk assessment there are two  
3 main components, and Dr. Potter was specifically addressing this  
4 earlier as well; how likely is the scenario in question to occur  
5 and how severe is the likely outcome once that scenario has  
6 occurred.

7 In the case of colony-level ecological models  
8 the former question relates really to sort of a dose response  
9 aspect of do we get doses out there and if the dose is out there  
10 does it get on a bee?

11 Where models actually help with that is kind  
12 of questionable because if we have the understanding about those  
13 routes of exposure, we can do a model that follows those but the  
14 routes are either basically there or not and its impact is  
15 either there or not. And we can have some sub-position models  
16 but, nah.

17 The latter question relates to how does those  
18 individual affects actually both interact together in one bee  
19 and how does that scale up into a society of bees and how is  
20 that society of bees scale to a population of bees and how does  
21 those population of bees provide the conservation and service  
22 endpoints that we have been discussing?

23 Those questions are a sweet spot for the  
24 ability of models. These relate to the impact of individual  
25 responses at sub-lethal effects. They can be ways to study the  
26 concomitant effects of different pathways. And we were talking



1 about I think you guys call them brews of pesticide before or  
2 something like that - mixtures, unintended or un-marketed  
3 mixtures of pesticide. This is a great place to try and explore  
4 those things.

5 For risk assessment itself and the models here  
6 presented that were described in the White Paper - and if I fail  
7 to mention it I'm sorry - I agree the review of the simulation  
8 models in the White Paper was wonderfully done. And as you guys  
9 said not completely exhaustive but representative and well  
10 described so thank you for doing that, that made my comments  
11 able to back up to say and analytic instead of just, oh do not  
12 do that with simulation.

13 So as those model presented were not  
14 specifically designed with these types of questions in mind,  
15 they do not specifically address these particular questions of  
16 risk assessment directly and in useful ways; but they could be  
17 very important in formulating answers within each level of  
18 concern to produce a globally useful risk assessment. So this  
19 is where that modularity that we were talking about earlier  
20 comes into play.

21 Well how do we understand the impact of this  
22 kind of toxicity to queen fecundity? Or to the dispersion of  
23 queen (inaudible) in a colony or to the ability of foragers to  
24 get home? Those are specific questions and specific effects  
25 that could be plug modularly into an overall model.

26 I would like to reiterate again that I do not



1 mean to suggest that we should plug in every available module  
2 because then both analytic and simulation models become unwieldy  
3 and the interpretation is very very difficult.

4 Again I will point to the analytic side of  
5 things to both designate and integrate which of these specific  
6 modules will be of most impact to the risk assessment outcomes  
7 for particular questions.

8 And again by employing those more algebraic  
9 analytic models it is going to be easier to assess which of the  
10 more specific fine-grain models or simulations require then  
11 their own empirical metrics to parameterize correctly and how to  
12 validate those by as I was saying sort of incidental validation  
13 portions of models.

14 I can give an example but I am going to leave  
15 it to Dr. James and others to call me on it if an example feels  
16 necessary for this question.

17 These sorts of models both analytic and  
18 simulation can generate insight into which valid threshold of  
19 outcome for risk assessment are going to be meaningful  
20 measureable endpoints for compromising the goals of all of the  
21 services we are talking about, both to ourselves and to bees  
22 themselves.

23 **DR. ROSALIND JAMES:** So the question asks  
24 which of the most important elements to consider in a model and  
25 I will add to what was said already. My view might be a little  
26 bit different than yours but for both theoretical and simulation



1 models there needs to be some sort of empirical validation  
2 before you use them.

3 And ideally I think for simulation models  
4 there really needs to have been done, or you can do it, some  
5 sort of sensitivity analysis, meaning that each component and  
6 how sensitive is that component to the outcome of the model.  
7 And then that tells you something about how accurate the data,  
8 how precise the data has to be that are the inputs.

9 And then of course hopefully the model gives  
10 you the critical elements that you are looking for as an  
11 endpoint, which I would think one of those would be to help  
12 identify what endpoints you need to request data from the  
13 registrant for.

14 **DR. THOMAS POTTER:** I generally agree with  
15 what Dr. Fefferman and also Dr. James has stated, I just simply  
16 want to add that there is perhaps another question and this is  
17 in direct response to Dr. Fefferman that we would ask about a  
18 model.

19 And that is, and again there are certain  
20 conditions that you have to apply to this, once calibrated  
21 and/or validated can it be used in the absence of additional  
22 empirical data to inform decisions. And certainly that would be  
23 critically important to the agency when we look particularly at  
24 Tier II and Tier III studies recognizing they are expensive and  
25 that there is a limited number of studies that can be done.

26 Is it possible to use data from a study that



1 was say conducted in Illinois to inform decisions about whether  
2 or not it is suitable to use the active ingredient in the same  
3 way in the state of Georgia for example?

4 So certainly these are critical questions that  
5 one would ask about a model in terms of what it can do and how  
6 effective it is as a tool because I see it as part of a toolkit  
7 that we use in risk assessment rather than maybe the other way  
8 around, which is the model is used to simply evaluate risk.

9 And I will sum up by saying and again maybe  
10 because I'm sitting in the exposure side of the room here I  
11 strongly think that considerably more work can be done and  
12 substantial progress can be made on the exposure side, which I  
13 think will serve to support particularly the Tier I testing and  
14 advance the process.

15 **DR. DAVID TARPY:** I echo the comments by the  
16 previous discussants.

17 **DR. DANIEL SCHLENK:** Other panel members.

18 **MR. JENS PISTORIUS:** As the last addition I  
19 concur to all what has been said. When mentioning to the model  
20 of Henry (phonetic) and specifically because that has caused a  
21 lot of trouble and concern and it was a comment I made to the  
22 previous answer.

23 I just wanted to point out that was not my  
24 work who found that this model in specific had was not validated  
25 for the purpose because there was one parameter and maximum  
26 daily rate of production of new work, which was not empirically



1 based.

2 And this is basically the work of Dr. Creswell  
3 (phonetic), and he made a technical letter to science, I do not  
4 know if it is published yet or not but I have it; it is  
5 confidential and it is his work. I want to point this out.

6 And this leads me to my part of the answer, I  
7 think considering the high variability, I am not quite sure if  
8 it is the same in the U.S., but in European region for instance  
9 for colonies to develop it is just important to point out that  
10 all of these parameters that are measured and that come into  
11 those models actually suits also the geographical location and  
12 the climate conditions.

13 For instance, colony growth is a completely  
14 different issue. When you have southern Europe, you have  
15 basically the colony growth is like this. There is a higher  
16 population in winter and it grows slower in spring and does not  
17 achieve such a growth rate basically. And then it goes to a  
18 moderate amount of bees. But what they do in Germany is they  
19 come from a very small number amount of bees and then there is  
20 basically explosion of the colony. You can call it a really  
21 massive increase of number of bees. And then in autumn there is  
22 a very big decrease.

23 And when those models are suitably adapted, I  
24 am very happy.

25 **DR. NINA FEFFERMAN:** I want to thank Mr.  
26 Pistorius for providing a fantastic example of what an analytic



1 model can show you, what kinds of colony growth trajectories you  
2 could expect, what are the parameters to change without having  
3 to go measure all of the parameters. Awesome, thank you.

4 **DR. DANIEL SCHLENK:** Any other comments?

5 **DR. ROSALIND JAMES:** I will add one more that  
6 I forgot to say. On the review of models you have the de grande  
7 (phonetic) bee pop model, but the newer varroa pop model is  
8 probably better and includes climate data in it also.

9 **DR. JEFF PETTIS:** This was a follow up to  
10 that. The only problem with using simulation models solely for  
11 that is that a northern researcher would never get to work in a  
12 southern climate, so that would be a drawback.

13 **DR. NINA FEFFERMAN:** For simulation I agree.  
14 For analytic, um.

15 **DR. DANIEL SCHLENK:** Any other comments from  
16 the panel? Okay, let me go to the agency; any questions of  
17 clarification?

18 **MR. KEITH SAPPINGTON:** I think we are good,  
19 thank you.

20 **MR. STEVEN BRADBURY:** I think it compliments  
21 what you all are talking about to the extent it can get captured  
22 in the report could be helpful.

23 So in my mind the Tier II or Tier III studies  
24 are physical models trying to provide insights into the real  
25 world, whatever the real world is. But you can only do a Tier  
26 II or Tier III study in certain places at a certain point in



1 time with that batch of bees. And either implicitly or  
2 explicitly we are running a model in our brain to project what  
3 that means in many places and at many times with different races  
4 of bees or whatever. And so those empirical models have  
5 shortcomings. I mean in empirical like those physical models.

6  
7 But all of those things are rolling through  
8 our heads and then the simulation models or the analytic models  
9 are ways to try to think about all of those things rolling  
10 around in our heads and using mathematics and other kinds of  
11 knowledge to try to help us think through that.

12 And none of them are right but all of them  
13 give us different insight and they are all at various stages of  
14 development.

15 So to the extent that question 13 and 14  
16 emerge in your writing, if there is any way that you can opine  
17 or provide advice on how you try to blend what we know today  
18 between insight of simulation or an analytic model might provide  
19 to help us be more explicit in the assumptions that we are going  
20 to use when we look at those physical models to make projections  
21 about what is happening in the southeast or what is happening in  
22 the northwest of the United States or the parries in Canada, the  
23 Sacramento watershed in California.

24 Because there is probably a blending of all of  
25 these, those physical models and what we do not really get real  
26 clear about what we are assuming when we look at the results of



1 physical models and how these analytical or simulation models  
2 can help inform what we should be saying about our assumptions.

3 I think that would just be helpful if  
4 something between 13 and 14 could try to blend what I think I  
5 heard in a lot of the comments but if you guys could bring it  
6 together that would, I think, help a lot of people not just us.  
7 Thanks.

8 **DR. DANIEL SCHLENK:** Okay, at this point this  
9 will be our final sort of commenting period so I would like to  
10 just go around the panel, if you have any final comments you  
11 would like to get on the record this is the time to do so. And  
12 begin with Dr. Sandy.

13 **DR. MARTHA SANDY:** I do not have anything to  
14 add.

15 **DR. STEPHEN KLAINE:** I for one would just like  
16 to thank the EPA for the document; it was great, and my bee  
17 colleagues for educating me about bees. This is a really  
18 important and very fascinating subject and I really look forward  
19 to see how this thing emerges. It has become obvious to me that  
20 we are not dealing with single bees to protect but we are  
21 dealing with colonies of bees, at least for honey bees.

22 So I think that is going to require some sort  
23 of out of the box thinking as we move forward. Concepts like  
24 homeostasis might be better applied to bee colonies than to  
25 bees. So anyways I just think it has been an excellent week and  
26 very informative.



1                   **DR. MARTHA SANDY:** If I could have a go back I  
2 would also like to say I really enjoyed meeting all of you and  
3 enjoyed the White Paper too to the EPA staff, thank you.

4                   **DR. JAMES MCMANAMAN:** I echo Dr. Sandy and Dr.  
5 Klaine's comments and appreciation of the White Paper and all  
6 the work that has gone into this and the discussions of the  
7 panel.

8                   I think the one thing that I came away from  
9 this was that, and it emphasizes that I do not think -- I think  
10 it was a clear indication that survival is not the only endpoint  
11 that should be considered and that should be taken to heart by  
12 the agency in terms of looking at other ways of evaluating the  
13 effects of pesticides.

14                  **DR. KENNETH DELCLOS:** I will just say it was a  
15 most interesting week but I have nothing to add here.

16                  **DR. DAVID TARPY:** I would like to echo all of  
17 those same comments. I think this has been a fantastic  
18 discussion, one that has been needed and I look forward to the  
19 evolution of this process.

20                  I hope everybody has gained an appreciation  
21 for just how messy and dirty honey bee colonies can be. And I  
22 think that that does not distract at all from their importance  
23 to our commercial production agriculture and just what an  
24 important issue this really is for our entire system.

25                  **DR. PAUL SCHWAB:** This has been quite an  
26 education for me. I did not have very much information about



1 bees before I started reading this White Paper. So my learning  
2 curve has been very steep but I appreciate the patience that all  
3 of you have express when I have been inclined to weave my way  
4 into this panel.

5 Just a little bit of bookkeeping, I did send -  
6 I have talked to Christina about this and to Tom - but I did  
7 send in a comment dealing with a correction to the absorption  
8 equation that was being used for contamination of puddles during  
9 a spraying event.

10 I know that we were supposed to have all of  
11 this into the record or it really could not officially exist so  
12 that is the point of this.

13 **DR. THOMAS POTTER:** I would like to thank  
14 first off the EPA staff that put together the White Paper,  
15 obviously a huge amount of work and probably done on a  
16 compressed timeframe I would expect. It was certainly  
17 informative and I can say I learned a tremendous amount from  
18 reviewing it and really enjoyed being part of the panel.

19 And I will say that I think that the outcome  
20 of this is certainly going to have a very strong impact, I  
21 think, on the whole process of ecological risk assessment. I  
22 think there are many core elements of this that will transcend  
23 into the whole area of looking at ecological risk.

24 **MR. JENS PISTORIUS:** First one technical  
25 comment to the bee issues we are talking about before and then  
26 on to the rest. I have been making a list with all the points,



1 you know, where I thought during the discussions I hoped this  
2 was going to be mentioned.

3 One last comment I have for synergistic  
4 effects for spray application on flowers and crops; when you  
5 know that there are a certain -- and I know that it is taken  
6 into consideration, I just want to point it out again -- that  
7 there are synergistic actions between two classes of compounds  
8 like the pyrethroids and the EBI fungicides. Please consider  
9 those synergisms as well as all others really documented. We do  
10 not have to test every sector, but with those two classes  
11 pyrethroids and EBI fungicides.

12 Please also do a higher tier risk assessment  
13 or do risk mitigation measures that are appropriate because we  
14 know we had problems with it in the past. But I think this is  
15 taken into consideration so I'm going to end it with a technical  
16 comment.

17 But now let me say a few personal words on  
18 this whole issue. Before I met the first EPA staff in 2011 at  
19 the SETAC Pellston conference, I will tell you frankly that one  
20 person told me what is going to happen. You are going to the  
21 U.S. and they'll let you talk, let you know your experiences and  
22 then they will say, thank you, and not take anything of this  
23 into account.

24 And I am deeply impressed, seriously, deeply  
25 impressed and I will go to this person and I will tell him all  
26 of my experiences. Really it is such a great honor to be here.



1 I was really impressed by all the good work. I have to thank  
2 the whole EPA staff. I think it is such an incredible effort  
3 that you have made.

4 This White Paper, when I read it I thought,  
5 hey this is great. Really, I think there is so much effort in  
6 this and I must say really I am deeply impressed how far you  
7 have brought this and how good the advancement is and we have  
8 been working together a little bit on other issues since 2011; I  
9 love to work with you. And it has been great fun, great honor  
10 and I think this has been such a great scientific progress.  
11 Thanks a lot, I take my hat.

12 **DR. JEFF PETTIS:** I would just like to offer  
13 thanks for the invitation to be part of the panel. I have very  
14 much enjoyed it. I would like to thank EPA for outlining the  
15 three protection goals, in particular, for me protection goal  
16 three which was broader than just honey bees. We tend to get  
17 caught up in honey bee biology and the complexities of that, but  
18 pollinated declined is bigger than just honey bee issues.

19 So I thank the effort put into the White  
20 Paper, in particular the protection goals I thought were spot  
21 on. Thank you.

22 **DR. NANCY OSTIGUY:** This was probably the most  
23 intense, but also the most fun four days that I have spent in a  
24 long time. Everyone has been incredibly collegial. It has been  
25 very informative. I have enjoyed myself thoroughly. I have  
26 collected also an amazing amount of information I am going to



1 take back to my students.

2 I have always been a defender of EPA because I  
3 think you get picked on too much but I have some very personal  
4 stories now to tell my students about what a wonderful job you  
5 are doing under very difficult circumstances.

6 And I also have to say something in defense of  
7 those messy bees. They are no messier to think about than when  
8 you think about human as a population. I feel extremely sorry  
9 for the sociologists that are trying to study us. And in many  
10 ways maybe we should think of ourselves as sociologists of honey  
11 bees.

12 But in defense of those bees they are  
13 extremely neat inside those colonies.

14 **DR. ROSALIND JAMES:** I almost want to say, I  
15 concur with my colleagues because I guess I will say the same  
16 thing. I was very glad to see EPA take on this issue. Everyone  
17 has been complimentary; I do not want to be too uncomplimentary  
18 but I do want to say, finally. It is a little behind times but  
19 I am very glad to see EPA take it on.

20 And when I started reading the White Paper I  
21 was very impressed. I mean throughout it. It was better than I  
22 expected I guess is what I mean. And I appreciate being invited  
23 to this panel with my beyond Apis mellifera views. Thank you.

24 **DR. GREG HUNT:** I concur; and I also am having  
25 my students read some papers that relates to these issues and we  
26 will be talking about it next week.



1                   And I thank the EPA for their efforts. I  
2 think that the increase emphasis in Tier I that is put in the  
3 White Paper is very good and I hope that we as a panel can  
4 provide a kind of a tiered response in our writings so that we  
5 can give you some, putting the simple things at the top and then  
6 the more future things at the bottom.

7                   **DR. NINA FEFFERMAN:** Thank you guys, all of  
8 you, the EPA members, the panel the public commenters, you guys  
9 running this, oh my God.

10                  I have had an awesome time and I have learned  
11 a huge amount and I have made friends and I like it. As I read  
12 into the existence record I want to say that there are some  
13 analytic models of honey bee colonies and I will provide some of  
14 those for the notes. Thank you.

15                  **DR. MAY BERENBAUM:** And to come crashing down  
16 on an incredibly mundane technical note just to read it into the  
17 record but I will follow that with some more inspirational  
18 language.

19                  Our discussions of prolonging the endpoint of  
20 Tier I larvae assay through pupation lacked literature  
21 documentation. There were some hand waving or remarks about,  
22 yes papers are out there. Well one such paper is "Honey Bee  
23 Risk Assessment: New Approaches for In Vitro larvae Rearing and  
24 Data Analyses" by Hendriksma, et al. "Methods in Ecology and  
25 Evolution" 2011. It is sort of a modification of Aupinel et al,  
26 but the point is it is a non-grafting method and the endpoint is



1 pre-pupation and it was used with some success.

2 On a higher note, as I mentioned 117 years  
3 inadvertent bee mortality caused by pesticide is really -- I am  
4 deliriously happy that it is now a national priority to protect  
5 not just honey bees but bees in general and I do not know if  
6 people will appreciate what a massive societal change that is.

7 Insect toxicology for decades involved how to  
8 kill insects so this is really un-trodden ground for the most  
9 part. And having said that the repeated references to data  
10 gaps, I think, obscures the fact that the White Paper represent  
11 there has been a tremendous explosion of extremely informative  
12 and relevant data so that we now can make science-based  
13 decisions on the best way to assess risk and protect bees.

14 So I am glad bees are having their day here in  
15 Washington D.C. and thank you for letting me be part of it.

16 **DR. DANIEL SCHLENK:** Let me say you are all  
17 making history. As I mentioned before this was a record number  
18 of questions that we have ever gotten, and you did it. You did  
19 do it. I want to congratulate you. This has been one of the  
20 best panels I have had the honor of being a part of.

21 I appreciate the tremendous hard work that  
22 many of you did throughout this week and days leading up to it  
23 of course. But also just to your collegiality and working  
24 together and being efficient in terms of answering the  
25 questions; that was just phenomenal. Great job, hat off to you  
26 guys.



1                   **DR. JEFF PETTIS:** A point about your  
2 leadership on the panel. There were some issues when you called  
3 it yesterday at 4:30, and we were like "why not press on?" and  
4 so there was a lot of discussion afterward but you made the  
5 right call there. We never would have gotten through these last  
6 questions with the level of detail and discussion that was  
7 needed. So you made a great call yesterday. We were pretty  
8 burnt yesterday.

9                   **DR. DANIEL SCHLENK:** Thanks, actually that was  
10 Dr. Jenkins pushing me on the right side over there. I wish I  
11 could take credit for that but.

12                   **DR. NINA FEFFERMAN:** I also just quickly want  
13 to acknowledge the amazing staff that has been helping us  
14 throughout; they have made this run smoothly as well and I want  
15 to thank them.

16                   **DR. DANIEL SCHLENK:** Definitely, where would  
17 we be without the SAP staff? Thank you very much, getting you  
18 all here and out safely and soundly hopefully.

19                   Again, thanks to the panel members for your  
20 tremendous work. It has been a pleasure to be a part of this.  
21 Obviously the EPA for the presentation, I totally concur on the  
22 White Paper and one of the things I think you hear at least  
23 among the academics is, even though you never what happens in  
24 terms of the policies that come out of this but in terms of  
25 academia and education the things that you are doing are making  
26 a difference at least in student's lives. Because I know every



1 single academic in here uses this information to educate our  
2 students. And that to me has always been a tremendous benefit  
3 of being part of these things; it is just so educational not  
4 just for me but for people that we also interact with.

5 So it is an exponential type of information  
6 sharing that comes out of this. So just congratulates you on  
7 that and again, I thought the White Paper was amazing. Not  
8 being familiar with this particular topic and the detail and  
9 clarity that was addressing these particular issues was just  
10 fabulous, kudos for that as well.

11 I would like to thank our public commenters,  
12 if they are still here, probably all gone but for their input  
13 and realism in terms of what this issue really means to a lot of  
14 those people.

15 EPA staff as well; I already said that. And  
16 with that I will turn it over to Dr. Bradbury usually have a  
17 word or two to say at the end.

18 **DR. STEVEN BRADBURY:** Real quick, just want to  
19 thank again the panel for all the hard work. We know we had a  
20 record setting number of questions and I appreciate all the work  
21 to do it. And every single topic you guys worked through I  
22 think gave us a lot of valuable insights. And the way you were  
23 synthesizing information like a super colony in action so we  
24 appreciate that too.

25 And we are committed to moving forward and I  
26 appreciate the comments about incremental steps and feedback



1 will help us move bit by bit and we will try to do better and  
2 better with our colleagues in Canada and California and around  
3 the world that we are all working on this.

4 I just want to thank you; you are a big  
5 contributor to advancing science and our ability to evaluate  
6 these situations.

7 **DR. FRED JENKINS:** I first want to start off  
8 by thanking Dr. Schlenk; this was a very important meeting,  
9 obviously a record breaking meeting and he did a great job in  
10 leading this panel through these charge questions.

11 And thank you so much to this panel. You all  
12 were fantastic and I appreciate you all agreeing to serve on  
13 this panel. When I send my invitation, I find you, send my  
14 invitations and talk to you and I really appreciate you all  
15 expertise, your talents, your commitments in this effort. And  
16 it has been truly a pleasure this whole week working with you  
17 all.

18 To the EPA-OPP team, there is a lot of hard  
19 work that goes on building up to having this meeting and OPP  
20 really works very hard. I used to work in OPP so I know and I  
21 used to work with EFED as a biologist, as an eco risk assessor  
22 for a long time. And so I know the work ethics in that office  
23 and I appreciate the opportunity to be there working with you  
24 all for this meeting.

25 Special thanks to Dr. Steven Bradbury the  
26 leader of the Office of Pesticide Programs. Dr. Donald Brady,



1 EFED Director, and the Branch Chief who actually led the team  
2 who did this work. Dr. Thomas Steeger, Thomas Moriarty, Mr.  
3 Keith Sappington, Christina Wendel, Kristina Garber, Reuben  
4 Baris, Joseph DeCant, all the speakers and scientists involved  
5 in developing this very good White Paper.

6 Thank you for the collaborative efforts of Cal  
7 EPA and PMRA, Ms. Mary Mitchell and Richard Bireley from Cal EPA  
8 Department of Pesticide Regulation. Thank you for your efforts  
9 in this whole process.

10 Thank you to all the public commenters and to  
11 all those who provided written public comments; there are a lot  
12 of written public comments in regulations.gov; so thank you to  
13 all those in the public for participating in this process.

14 And I would be of remiss if I personally did  
15 not thank my FIFRA SAP colleagues under the leadership of Laura  
16 Bailey, my colleagues Joseph Bailey, Sharlene Matten, Joyce  
17 Coates, Lu-Ann Kleibacker, our team lead, and Shirley Percival.

18  
19 The meeting report will be available no later  
20 than 90 days after the close of this meeting. This meeting is  
21 now officially concluded. Everyone have a nice weekend and  
22 thank you all.

23 (WHEREUPON the meeting was adjourned)  
24  
25  
26



FIFRA SCIENTIFIC ADVISORY PANEL (SAP)

OPEN MEETING

POLLINATOR RISK ASSESSMENT FRAMEWORK

DOCKET NUMBER: EPA-HQ-OPP-2012-0543

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