MEETING

STATE OF CALIFORNIA

OFFICE OF ENVIRONMENTAL HEALTH HAZARD ASSESSMENT

PROPOSITION 65

DEVELOPMENTAL AND REPRODUCTIVE TOXICANT

IDENTIFICATION COMMITTEE

JOE SERNA JR.

CALEPA HEADQUARTERS BUILDING

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COASTAL HEARING ROOM

SACRAMENTO, CALIFORNIA

THURSDAY, MAY 7, 2015

10:04 A.M.

JAMES F. PETERS, CSR
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A P P E A R A N C E S

COMMITTEE MEMBERS:

Ellen B. Gold, Ph.D., Chairperson

Diana Auyeung-Kim, Ph.D.

Laurence Baskin, M.D.

Suzan Carmichael, Ph.D.

Ulrike Luderer, M.D., Ph.D.

Isaac Pessah, Ph.D.

Charles Plopper, Ph.D.

STAFF:

Dr. Lauren Zeise, Acting Director

Ms. Carol Monahan Cummings, Chief Counsel

Dr. James Donald, Chief, Reproductive Toxicology and Epidemiology Section

Mr. Sam Delson, Deputy Director

Mr. Mario Fernandez, Staff Counsel

Ms. Fran Kammerer, Staff Counsel

Dr. Farla Kaufman

Dr. Melanie Marty, Assistant Deputy Director, Scientific Affairs Division

Dr. Martha Sandy, Chief, Reproductive and Cancer Hazard Assessment Branch

Dr. Lily Wu, Reproductive and Cancer Hazard Assessment Branch

A P P E A R A N C E S C O N T I N U E D

ALSO PRESENT:

Mr. Bill Allayaud, Environmental Working Group

Mr. Robert Chadwick, Can Manufacturers Institute

Dr. Julie Goodman, American Chemistry Council

Ms. Ann Grimaldi, Art and Creative Materials Institute

Dr. Steven Hentges, American Chemistry Council

Mr. Avinash Kar, Natural Resources Defense Council

Dr. Beth Mileson, Technology Sciences Group

Dr. Jay Murray, American Chemistry Council

Ms. Emily Reuman, Breast Cancer Fund

Dr. Johanna Rochester, TEDX

Mr. Brian Rodriguez, Center for Environmental Health

Mr. John Rose, NAMPA

Ms. Gretchen Lee Salter

Dr. Anthony Scialli, American Chemistry Council

Ms. Renee Sharp, Environmental Working Group

Dr. Veena Singla, Natural Resources Defense Council

Dr. Tasha Stoiber, Environmental Working Group

Dr. Rebecca Sutton, San Francisco Estuary Institute

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P R O C E E D I N G S

CHIEF COUNSEL MONAHAN CUMMINGS: Good morning. My name is Carol Monahan Cummings. I'm the Chief Counsel for the Office of Environmental Health Hazard Assessment. Before we get started with our regular meeting today, I need to make a short announcement.

Sadly, I have some bad news for you all. Dr. George -- Dr. Alexeeff...

ACTING DIRECTOR ZEISE: Carol, why don't I do this or Mario will.

CHIEF COUNSEL MONAHAN CUMMINGS: All right. Mario will come up.

STAFF COUNSEL FERNANDEZ: Okay. Dr. George Alexeeff, Director of the Office for the last four years or so is seriously ill and is not expected to return. Many of you have known Dr. Alexeeff for many years and you can appreciate how much he'll be missed. Fortunately, the Office is in good hands. Dr. Lauren Zeise, who's been Deputy Director for Scientific Affairs and is well known to many of you, was appointed to the -- by the Governor as Acting Director on Monday. She'll be participating in the meeting today in that capacity.

With that, I'll turn the meeting over to Dr. Zeise and Carol Monahan Cummings will be speaking later on.

ACTING DIRECTOR ZEISE: Thanks, Mario. I'm sorry for you all to hear the news this way.

Today, we have one major agenda item in front of us, and that is to look at the female reproductive toxicity of bisphenol A. There's a very large database for bisphenol A. And so in the event that -- where the panel is unable to work through all of that evidence and sort through it all, we do have a second meeting scheduled on May 21st.

I would like to welcome the Committee and the audience to this meeting. And before we get started to the heart of the matter, just some housekeeping. First, this meeting is being transcribed and is being broadcast via webinar. So I want to remind people to speak directly into the microphones.

As far as logistics, if you go out this exit door and turn to the left, you'll see -- and walk down the hall, you'll find the restrooms and the drinking fountains. In the event of a fire alarm or any other reason to evacuate the room, please leave by the lighted exits at the back, take the steps down, and exit the building, and we'll relocate across the street.

We expect to be taking breaks during the meeting for the court reporter. And then lunch will be -- we'll take a more extended break for lunch. And the cafeteria

is downstairs.

Okay. So what we'll do now is introduce our committee members. First I'll introduce the existing members and then the new members. So at my far right is Dr. Laurence Baskin. He's the chief of pediatric urology and professor of urology and pediatrics. And he's a surgeon scientist at the University of California, San Francisco.

Get to Dr. Kim in a second.

Dr. Ulrike Luderer who is a professor of medicine in the School Medicine, UC Irvine. Next to me is -- my immediate right is Dr. Charles Plopper. He's professor emeritus -- oh, sorry. I'm going to get to you later.

(Laughter.)

ACTING DIRECTOR ZEISE: And then our Chair, to my immediate left, Dr. Ellen Gold, who is professor and chief, Division of Epidemiology in the Department of Public Health Sciences at UC Davis. Then to her left is Dr. Isaac Pessah, who's professor and Associate Dean of the School of Veterinary Medicine at UC Davis.

Okay. Now, for the new members. Next to Dr. Pessah is Dr. Suzan Carmichael. She's the associate professor, neonatal and developmental medicine at Stanford University. Dr. Carmichael is an epidemiologist who before coming to Stanford in 2010 held positions at the

March of Dimes Foundation, including division director of epidemiology.

Then to my right is Dr. Plopper, professor emeritus, Department of Anatomy, Physiology, and Cell Biology, UC Davis School of Veterinary Medicine. Dr. Plopper started his career with a Ph.D. in anatomy. And since coming to UC Davis in 1979 held positions in his department including Chair and professor.

And then Dr. Diana Kim is next to Dr. Luderer. She is Director of Toxicology at Allergan, Inc. Dr. Kim is a toxicologist who, before coming to Allergan in 2010, held several research positions at Charles River Laboratories including Associate Director of Research.

Now, I'd like to swear in our new members. If Dr. Plopper, Dr. Kim, and Dr. Carmichael, if you could please stand, and if you could raise your right hand and repeat after me.

I, and if each of you could say your name --

COMMITTEE MEMBERS: I --

ACTING DIRECTOR ZEISE: -- do solemnly swear --

COMMITTEE MEMBERS: -- do solemnly swear --

ACTING DIRECTOR ZEISE: -- that I will support and defend --

COMMITTEE MEMBERS: -- that I will support and defend --

ACTING DIRECTOR ZEISE: -- the Constitution of the United States --

COMMITTEE MEMBERS: -- the Constitution of the United States --

ACTING DIRECTOR ZEISE: -- and the Constitution of the State of California --

COMMITTEE MEMBERS: -- and the Constitution of the State of California --

ACTING DIRECTOR ZEISE: -- against all enemies, foreign and domestic --

COMMITTEE MEMBERS: -- against all enemies, foreign and domestic --

ACTING DIRECTOR ZEISE: -- that I will bear truth faith and allegiance --

COMMITTEE MEMBERS: -- that I will bear true faith and allegiance --

ACTING DIRECTOR ZEISE: To the Constitution of the United States --

COMMITTEE MEMBERS: -- to the Constitution of the United States --

ACTING DIRECTOR ZEISE: -- and the Constitution of the State of California --

COMMITTEE MEMBERS: And the Constitution of the State of California --

ACTING DIRECTOR ZEISE: -- that I take this

obligation freely --

COMMITTEE MEMBERS: -- that I take this obligation freely --

ACTING DIRECTOR ZEISE: -- without any mental reservation or purpose of evasion --

COMMITTEE MEMBERS: -- without any mental reservation or purpose of evasion --

ACTING DIRECTOR ZEISE: -- and that I will well and faithfully discharge --

COMMITTEE MEMBERS: -- and that I will well and faithfully discharge --

ACTING DIRECTOR ZEISE: -- the duties upon which I am about to enter.

COMMITTEE MEMBERS: -- the duties upon which I'm about to enter.

ACTING DIRECTOR ZEISE: So congratulations, and welcome.

So now I'd like to introduce our OEHHA -- staff of the Office of Environmental Health Hazard Assessment, OEHHA, our Chief Counsel, Carol Monahan Cummings, Martha Sandy who is the Branch Chief for the Reproductive and Cancer Hazard Assessment Branch, Dr. Melanie Marty, who's Assistant Deputy Director for Scientific Affairs. Maybe you should raise your hands as I walk through you, so the panel knows, because I am kind of jumping around.

Dr. James Donald, Section Chief for the Reproductive and -- sorry for the Reproductive Toxicology and Epidemiology Section. And then other staff in his section, Dr. Lily Wu and Dr. Farla Kaufman. Sam Delson, Deputy Director, External and Legislative Affairs. And then our Proposition 65 staff, Esther Barajas-Ochoa, and Monet Vela. Is Monet in the audience?

Okay. Now, Carol will make some introductory remarks.

CHIEF COUNSEL MONAHAN CUMMINGS: Good morning. I just wanted to also introduce Mario Fernandez, who's on my right. He's an Assistant Counsel with the Office. And he will be here in my absence if I have to leave the room.

I always give a little introduction for the staff -- or the Committee members given that you're only here once a year. I just wanted to remind you of a few things. In your binders and in the materials that we provided you before the meeting, you have the criteria that was adopted by the Committee that can provide guidance to you in terms of how to approach the scientific question that is before you today. Hopefully, you've had a chance to look at that. At any time if you need to take a break to review anything, including that criteria, just let the chair know that.

Your listing or not listing decision today should

be based on that criteria and not consideration of future impacts of a listing. For example, if you hear some comments about the effect of a warning for a particular product or exposure, that is not a question before the Committee and is not part of your consideration.

You will hear, when you are at the point of taking a -- making a decision, that there is a scientific standard that you need to determine whether or not it's been met. We call that the Clearly Shown Standard, but it will get repeated a number of times today. Just for your information, that is not a legal standard of proof. It's not something like beyond a reasonable doubt, which you can hear sometimes in court proceedings. What it is is it's a scientific standard and it's a judgment call that you are asked to make. It has a legal effect, but it isn't a question of a legal determination by this group.

Your Committee can decide to list a chemical based on animal evidence only. You're not required to determine that a chemical has been shown to be a human developmental or reproductive toxicant or whether or not human -- current human exposures to the chemical are sufficiently high enough to cause reproductive toxicity.

The members of this Committee were appointed by the Governor because of your scientific expertise. And you are not required nor you don't need to feel compelled

to go outside that charge. Also, in the event you feel you've had -- you have insufficient information or that you need more time to think about or discuss the question that's before you today - I know it's a very complex set of scientific information - there is no requirement that you make a decision today.

As you know, there is a meeting that's already been scheduled for May the 21st, the second day of the meeting, in the event that you need that time. And then there's also the opportunity to just say that you want the chemical brought back to you at another time with some additional information if you feel you need it.

So does anybody have questions in that regard?

All right. Feel free to ask me during the course of the meeting if you have questions.

Thank you.

ACTING DIRECTOR ZEISE: Okay. I will now turn the meeting over to Dr. Gold.

CHAIRPERSON GOLD: Thank you. Good morning. First of all, I want to thank the OEHHA staff and all the members of the Committee, as well as the public for all their hard work and effort. There is a voluminous set of Documents that have been before us. And so I know everybody has been working hard. So I just want to appreciate everyone's time and effort.

The other thing I would like so say in the interests of having an open and transparent process, we are allowing each member of the public five minutes, if they identified that they would like to say something, but it was also possible to request additional time. And we received advance requests from the ACC, the ACMI and the NRDC for additional time for a coordinated group presentation of their information from each of them. So they've been given the following time limits: The NRDC was given 15 minutes, the ACC 20, and the ACMI 20 minutes as well.

For any of you that want to make public comments, there will be blue cards, I believe, available in the back. And if you can give them to Esther, then at the time of the public presentations, we will acknowledge you and have you come up and give your presentation.

And I think, by way of introduction, that's all I have to say at this time. And I'm going to turn it over to Dr. Jim Donald who's going to do the staff presentation.

(Thereupon an overhead presentation was presented as follows.)

DR. DONALD: Thank you Dr. Gold.

I'm afraid allergies are trying to rob me of my voice, so I hope that I get through this without losing it

entirely.

Could I have the slides, please.

All right. Thank you.

For members who have participated at previous meetings, you'll note that there are going to be some differences in this meeting from our usual process. One difference is that, at this point, usually OEHHA staff have prepared technical summaries of all of the data before the Committee. And we would make a presentation on them at this stage. We have not done that for this meeting.

Instead, I've been asked to briefly review why BPA is before the Committee today for consideration for listing as causing reproductive toxicity, but being considered today solely on the basis of female reproductive toxicity.

--o0o--

DR. DONALD: So BPA has been considered previously by this Committee in July of 2009. The Committee considered whether BPA had been clearly shown by scientifically valid testing, according to generally accepted principles to cause reproductive toxicity. And at that time, the Committee considered all of the categories of reproductive toxicity, male reproductive, female reproductive, and developmental toxicity.

Based on the data available at that time, the Committee voted unanimously on all of those categories that BPA had not been clearly shown to cause reproductive toxicity. However, in the course of the meeting, the Committee specifically requested the opportunity to revisit consideration of bisphenol A, if additional epidemiological or other particular types of data on reproductive and developmental toxicity became available.

--o0o--

DR. DONALD: Materials provided in 2009 to the Committee, the hazard identification materials, were comprised primarily of four review documents. One was prepared by OEHHA. It provided an integrative evaluation and review of all of the relevant toxicity data, the relevant reproductive and developmental toxicity data. It provided information on pharmacokinetics and mechanistic data, and it also provided individual study summaries for studies that were not covered by any of the other review documents.

The second document was a monograph by the National Toxicology Program, Center for the Evaluation of Risk to Human Reproduction, which considered specifically the potential human and reproductive -- excuse me, human reproductive and developmental effects of bisphenol A. And that document was published in 2008.

The third review was a more general risk assessment conducted by the European Union on the toxicity of bisphenol A, and published in 2003. And the fourth review -- oops. The fourth review was an update of that risk assessment that was published in 2008. The final part of the hazard identification materials were all of the materials submitted to OEHHA and forwarded to the DART Identification Committee during a public -- a 60-day public comment period that preceded the meeting.

--o0o--

DR. DONALD: Okay. The reason why you're being asked today to consider only the female reproductive toxicity of bisphenol A is that OEHHA has determined that substantial new epidemiological and toxicological data on bisphenol A and its potential to cause female reproductive toxicity have become available since 2009. So consistent with the Committee's request to revisit it, we brought it back to you.

One example of that, one of the things that helped us reach that determination was a review published in 2014 by Peretz et al. in Environmental Health Perspectives. It provided a useful compilation of the relevant data. We're limiting consideration today only to female reproductive toxicity essentially for practical reasons. There is, as already alluded to, a considerable

volume and complexity to that data, so we wanted to give the Committee an opportunity to thoroughly and appropriately evaluate that endpoint. And that should not be interpreted to mean that other endpoints are not of concern.

The Committee may be asked to look at other endpoints, such as male reproductive toxicity, at future meetings.

--o0o--

DR. DONALD: So for this meeting, in addition to all of the materials that were provided to the Committee in 2009 and which have been provided to you, we've provided a substantial amount of additional information. This time, as I mentioned, OEHHA staff have not provided detailed summaries of the studies. Instead, we've only provided a general overview of the hazard identification materials.

Part of the materials you received was the review I already mentioned, published in Environmental Health Perspectives, that looked at bisphenol A and reproductive health and considered data published between 2007 and 2013. That's provided to you as a useful compilation and summary of the data. And in that vein too, we also provided you with the supplemental materials to that published paper that are available on-line and consist of

hopefully useful summary tables.

To help the Committee focus specifically on female reproductive toxicity, OEHHA staff went through that document, identified the sections that directly pertain to female reproductive toxicity, identified all of the articles cited in those sections, and we have provided you with copies of all of those articles.

We also conducted a literature search to update the materials with studies that had been published after the completion of the 2014 Environmental Health Perspectives review, and we've provided you with all of the relevant studies that we identified.

We went back to the 2009 hazard identification materials and did something similar with the four review documents that were provided at that time. We went through them, identified the sections specifically pertinent to female reproductive toxicity. And all of the articles and reports cited in those sections, we retrieved all of them that were available to us and provided them to you.

And the final part of the current hazard identification materials are the additional public comments and related materials that were submitted during the public comment period that preceded this meeting. And I'd just note that they did include some substantial

1 additional materials, such as the 2014 U.S. Food and Drug
2 Administration and 2015 European Food Safety Authority
3 safety assessments of bisphenol A looking specifically at
4 its safety in relation to human exposures resulting from
5 BPA's use in food packaging.
6 --o0o--
7 DR. DONALD: So I've already alluded to the
8 extent and complexity of the data on female reproductive
9 toxicity. We provided you in total with about 320 papers
10 and reports relevant to female reproductive -- or the
11 potential female reproductive toxicity of bisphenol A.
12 Two hundred ninety of those were cited in the five review
13 documents that I've mentioned, and 30 were papers that
14 were published subsequent to the most recent of those
15 reviews.
16 And I'll just note in passing that we found there
17 were 41 reports, relevant reports, cited in those reviews
18 that we were not able to attain, that were unavailable to
19 us, and so we could not obviously provide them to you.
20 --o0o--
21 DR. DONALD: In terms of the substantial increase
22 in relevant information, this is just an overview of the
23 studies identified in Peretz et al., categorized as they
24 categorized them. This is the number of studies that were
25 published after 2009, and so were not available to the

DART committee the last time it considered bisphenol A. And you'll note that in terms of additional epidemiologic data, there were 13 studies looking at female human reproductive outcomes, and eight studies looking at human pregnancy and birth outcomes, so a substantial increase in the epidemiologic data.

--o0o--

DR. DONALD: And additionally in the studies that OEHHA identified as being published after the Peretz et al. review, again the studies were focused on a range of relevant outcomes, but ten of them were also additional epidemiologic studies.

--o0o--

DR. DONALD: The last thing I've been asked to very briefly review is, since you're charged today with determining whether OEHHA -- whether BPA has been clearly shown by scientifically valid testing, according to generally accepted principles to cause female reproductive toxicity, what constitutes the generally accepted principles for identifying female reproductive toxicity.

Well, recognizing, of course, that there's always room for varying opinions, we look to publications that can be interpreted to reflect the generally accepted principles or the consensus opinion in this regard. One such publication is the U.S. EPA's Guidelines for

Reproductive Toxicity Risk Assessment, which identifies a range of endpoints that U.S. EPA considers to be female-specific endpoints of reproductive toxicity. And I would note that that document went through extensive public and peer review when it was being prepared and finally published, and so can reasonably be interpreted to represent the generally accepted principles.

Most of the endpoints are probably fairly self-evident. The condition of the reproductive organs in terms of weights and the condition by visual and histopathological examination, and those organs, of course, would include the ovary, the uterus, the vagina, but also the pituitary, the oviduct, and the mammary gland.

Effects on estrous and menstrual cycling can be indicative of female reproductive toxicity. Affects on sexual behaviors, both those that be can directly assessed, such as lordosis or time to mating in animal models or those assessed indirectly by measures such as presence of vaginal plugs or vaginal sperm in rodent models.

Changes in female sex hormones are obviously relevant, including effects on luteinizing hormone, follicle stimulating hormone, estrogen, progesterone, and prolactin.

Another consideration is affects on lactation, both in terms of the quantity and quality of milk produced, which can be assessed directly or -- again, indirect measures can include growth of suckling offspring. Early onset of reproductive senescence in females is clearly a relevant endpoint.

The last thing I'd direct your attention to is development of the female reproductive system is considered obviously a female reproductive metric, or metric of female reproductive toxicity. But obviously also, it can be considered a metric of developmental toxicity. And I'll come back to that point in a moment.

--o0o--

DR. DONALD: U.S. EPA guidelines were published in 1996, almost 20 years ago. So one consideration is even if those -- if they represent the generally accepted -- or represented the generally accepted principles, then do they still represent them?

An indication that they do is the relatively recent publication by the United Nations Globally Harmonized System of Classification and Listing of Chemicals, which identifies essentially the same list of endpoints of female reproductive toxicity as those identified by U.S. EPA 20 years ago.

Two things on this list though that I would draw

your attention to that were not on U.S. EPA's list of female specific endpoints are fertility and pregnancy outcomes.

--o0o--

DR. DONALD: The U.S. EPA identified those types of effects under their compilation of couple-mediated endpoints. And the points I wanted to make here is that you've been provided with data on pregnancy outcomes. Some of the outcomes -- the relevant pregnancy outcomes included in this list are fetal death rate, or fetal mortality, and fetal birth weights.

The reason why this -- it's important to consider this is that, as we know as biologists, reproductive toxicity -- or female reproductive toxicity does not exist in isolation from other types of toxicity. There may be clear evidence of reproductive toxicity where it is difficult or perhaps impossible to determine the contribution of effects on the female reproductive system, the effects on the male reproductive system and direct effects on the conceptus.

So in instances where pregnancy outcome is affected, it's important to consider that the possibility that the fetal development or the conceptus development was affected by the -- or was mediated throughout adverse effects of the chemical on the female reproductive system

impairing the ability of that reproductive system to maintain a healthy pregnancy.

So the overall message, I guess, is that it's very important to consider the entire scope of the data, both the empirical outcome data and the mechanistic data to determine -- and to integrate that information to determine how strong the evidence is that bisphenol A actually causes female reproductive toxicity.

So I will stop at this point and my colleagues who were introduced earlier, Dr. Wu and Dr. Kaufman, respectively are particular experts on the female reproductive system and on epidemiology. So with their help, I will be happy to try and address any questions you have at this point or at any time later in the meeting.

Thank you.

CHAIRPERSON GOLD: Thank you, Dr. Donald. Do either of you have anything additional that you want to add?

DR. WU: Not right now.

CHAIRPERSON GOLD: Okay. So, at this time, I first want to see if there are any questions from Panel members on the staff presentation?

Hearing none.

Our next item on the agenda is turn to the Committee's discussion of the material that we've been

given. And we divided this up among the Committee members and tried to have a primary discussant and a secondary discussant. And we divided it up largely by species, but also a little bit by topic area.

So we'll start with discussion of the rodent publications. And Dr. Luderer is going to lead that followed by Dr. Pessah. Let me just say that following that we'll deal with non-human primates and Dr. Auyeung-Kim will lead us on that and Dr. Plopper will be secondary. And then we'll deal with human data, and Dr. Carmichael will lead that, and I will follow-up. And then finally, we'll deal with androgen steroidogenesis and exposures in females that affect males, and Dr. Baskin will lead us on that.

So without further ado, I'm going to turn it over to Dr. Luderer.

COMMITTEE MEMBER LUDERER: Thank you, Dr. Gold. As has already been mentioned, there have been -- there was a voluminous volume of studies given to the Committee to review. What I'm going to do is to focus primarily on the studies that have been published since the last DART meeting in 2009, so -- and the rodent studies since that last DART meeting have added particularly, I believe, to the weight of evidence that female reproductive toxicity is caused by early life exposures to bisphenol A. And so

1 the majority of the endpoints that I'll be talking about
2 relate to early life exposures.
3 So I'm going to start by talking about two high
4 quality recent studies that were published in 2013 and
5 2014. These studies had multiple doses spanning orders of
6 magnitude administered -- of BPA administered perinatally
7 to rats, and both studies reported statistically robust
8 effects on different sexual development endpoints.
9 One of these was Christiansen et al, from 2014.
10 And this study used pregnant Wistar rats, which is a
11 sensitive strain, dosed orally by gavage from gestational
12 day 7 to 22 -- postnatal day 22 with 0, 0.25 -- 0.025,
13 0.25, 5 or 50 milligrams per kilogram per day bisphenol A,
14 BPA, covering the sensitive windows for reproductive
15 system development. And this study was sufficiently
16 powered to detect differences in the endpoints examined.
17 Attention was paid and I'll discuss this briefly
18 for these first two studies and make comments when I talk
19 about other studies.
20 Cages and water bottles were not polycarbonate.
21 There were polysulfone to minimize potential BPA exposure
22 from that source. The feed was phytoestrogen or at least
23 soy and alfalfa free. There was a single skilled
24 technician blinded to exposure who measured AGD,
25 anogenital distance. And BPA concentrations in dosing

solutions were confirmed.

So in this study anogenital distance was significantly decreased in females at all BPA dose groups relative to controls. And the investigators also compared the controls in this study to two other recent studies in the same strain by their group and they found no difference in the controls, eliminating the possibility that unusually high values in the controls might have explained their findings.

There were no effects on ovarian weights examined at postnatal day 16. So this study shows effects on female anogenital distance at birth, with prenatal and post -- prenatal exposure to bisphenol A, which is indicative of altered endocrine signaling during development and may be associated with altered reproductive function later in life.

McCaffrey et al, in 2013, performed another high quality study that examined the impact of early life exposure on sexual differentiation of two sexually dimorphic brain areas, the anteroventral periventricular nucleus, or AVPV, and the sexually dimorphic nucleus of the preoptic area or the SDN-POA. Both of these are hypothalamic nuclei. The former is identified by tyrosine hydroxylase positive dopaminergic neurons and the latter by calbindin positive neurons.

Males and females importantly start out with the same number of neurons in both of these nuclei, but then estradiol signaling via estrogen receptor alpha is thought to have opposite effects on cell death in the two nuclei, so that in males the SDN-POA is bigger while the AVPV is bigger in females.

So this study used Pregnant Long Evans rats, another sensitive strain dosed with 0, 10, 100, 1000, and 10,000 micrograms per kilogram per day in corn oil orally in a cookie or with 17beta estradiol included as a positive control from gestational day 12 to postnatal day 10.

In this study, there was a main effect of sex on both regions, and there was a main effect of bisphenol A, but no interaction between BPA and sex on the AVPV, so I'm going to highlight that. The tyrosine hydroxylase immunoreactive neurons in both females and males were significantly decreased in number, so that in the females, the females were masculinized compared to the respective control females in all groups, except for the 1000 microgram per kilogram group which approached significance and there were also decreased in males. There were also significant effects on the SDN-POA and the calbindin immunoreactive cells in that nucleus in males, but not in females.

So this study shows clear effects of BPA dosing on brain sexual differentiation in females, with masculinization of the AVPV. And this could potentially affect timing of puberty and ability to have normal preovulatory LH surges.

So in addition to these two high quality recent studies, I think there are key endpoints for which there have been multiple in vivo studies of varying quality, often supported by in vitro studies. And some of these studies were published at the time of the last DART review of BPA and were summarized in the 2009 DART document, and I'll just highlight a few of those. And additional studies since then have added to evidence concerning affects of BPA on these endpoints.

And these endpoints include meiosis errors, oocyte cyst breakdown and primordial follicle assembly, lesions of the ovaries, oviducts and uterus, and alterations in mammary gland development and hyperplasia/neoplasia of mammary glands following early life exposure.

So regarding the meiosis errors, I'll summarize some of those studies first. Numerous studies in several species have examined the effects of early life bisphenol A exposure on meiosis progression in females.

One of the first studies was the Hunt et al.

study from 2003, which reported that oral pipet dosing of C57 Black 6 mice with 20, 40, or 100 microgram per kilogram per day from postnatal day 21 to 28, so just after weaning but prior to puberty, caused -- that was after -- with 7 days of dosing -- caused dose-dependent and time-dependent at the 20 nanogram per kilogram BPA doses was only tested in the time-dependent study after 7 days, but not after 3 or 5 days. Increases in congression failures at meiosis -- at metaphase II and that is failure of chromosomes to properly align on spindle) in germinal vesicle oocytes that were collected from antral follicles, cultured overnight and then examined if they extruded a polar body. This was a good quality study with multiple thing -- blinding, details provided about cages, water bottles, et cetera.

Eichenlaub-Ritter et al. in 2008 conducted a similar study in mice of the C57 Black 6 times CBA/Ca F1 strain. They also dosed for the same dosing interval with oral doses at the same dose, but they did dosing by gavage rather than pipet. They also collected germinal vesicle stage oocytes cultured overnight, but they did not observe congression failures or other significant meiotic abnormalities. They did, however, observe meiotic arrest with failure to emit a polar body and increased bivalent chromosomes and polyploidy when the germinal vesicle

oocytes were matured in vitro with bisphenol A at a concentration at 43 micromolar, but not lower concentrations.

So differences between these two studies have been widely discussed. And some of the differences that might explain the divergent results include the oral dosing method, gavage versus pipet dosing, different diets provided to the mice in different strains.

The Eichenlaub-Ritter group also published two supportive studies that were done on cultured follicles. In the Lenie et al. study from 2008, they cultured secondary follicles for 12 days to the preovulatory stage and ovulated them, and they found arrest at the germinal vesicle breakdown stage with failure of polar body extrusion only in the highest 30,000 nanomolar group.

However, metaphase II abnormalities consisting of unaligned chromosomes abnormal spindles were observed also at lower concentrations of 3, 30, 300 nanomolar, as well as 3000 nanomolar and they were more severe at the lower concentrations of bisphenol A. The same group used a similar culture paradigm with 0, 3, and 300 nanomolar concentrations and measured and found allele hypomethylation errors of maternally imprinted genes and decreased histone 3K9 trimethylation with the 3 nanomolar dose.

A couple of other -- some other studies that have been done used one -- the next study that I'll talk about Chao et al. from 2012 used two earlier postnatal dosing intervals in CD-1 mice injected BPA subcutaneously in saline. In the first experiment they injected 0, 20, or 40 micrograms per kilogram per day from postnatal day 7 to 14, and sacrificed the next day. In the second experiment, they used the same route and doses, but every five days, postnatal day 5, 10, 15, and 20 with sacrifice on day 21.

I'm going to talk first just here about the meiosis endpoints that they looked at, which was only in the second experiment. They collected oocytes from antral follicles, matured them in vitro for 16 hours and scored for maturation. They observed decreased percentages of oocytes with germinal vesicle breakdown in the highest dose groups of 40 microgram per kilogram per day relative to control, but no differences in percentages with the first polar body extruded.

And they reported increased spindle abnormalities in BPA exposed oocytes at MI. For the oocytes that did reach MII, however, they didn't observe any spindle abnormalities.

They also reported decreased -- dose-dependently decreased methylation of two maternally imprinted genes

again, but not paternally imprinted genes, and decreased expression of several DNA methyltransferases.

This study was -- had several flaws with insufficient detail about some of the experimental methods. However, the study does support that early postnatal treatment with BPA affects meiosis I.

So the next two studies affecting -- regarding meiosis used a prenatal rather than a postnatal dosing window and they examined effects on meiosis also.

So the Susiarjo et al. study from 2008, in that study they treated C57 Black 6 mice with subcutaneous pellets releasing 20 microgram per kilogram per day bisphenol A from gestational day 11.5 to gestational day 18, and then prepared chromosomal spreads for -- to examine MI. And some females were then also treated the same way and sacrificed at 4 to 5 weeks of age for germinal vesicle oocyte collection, analyses of MI oocytes after 1 to 2 hours maturation, and MII after 16 hours or they superovulated some females, mated them, and examined the cleavage stage embryos.

So in this study, they observed no differences in the percentage of oocytes at prepachytene, pachytene or diplotene of meiosis I at gestational day 18, but they did observe synaptic abnormalities consisting of incomplete synapses and end-to-end associations of nonhomologous

chromosomes. They also observed increased recombination foci and altered distributions of the foci along the chromosomes using two different methods, one on pachytene oocytes from the fetal ovaries, and then again from the metaphase I spreads on oocytes collected at 4 to 5 weeks of age. They also observed statistically significantly increased aneuploidy on metaphase II spreads and nonsignificantly increased aneuploidy in the two cell embryos.

Finally, regarding the meiosis endpoints, Zhang et al. in 2012 dosed CD-1 mice by oral pipet with 20, 40, or 80 microgram per kilogram per day bisphenol A in a similar dosing window, 12.5 to 18.5 days post coitum. And they used only that highest BPA dose to examine meiotic progression, the 80 microgram per kilogram, and observed delayed meiotic progression between 15.5 and 19.5 dpc's, with decreased percentages of oocytes from BPA-treated mice reaching zygotene by day 15.5, pachytene by 17.5, and diplotene by day 19.5. And this was associated with significantly decreased expression of the meiosis initiator Stra8 at 17.5 dpc.

So this -- again this study had several -- lacked several details, such as about the vehicle, the diet, the N per group. However, again it adds I think to the weight of the evidence that early life exposure in this case

gestational BPA affects meiosis in females.

So overall, the studies in which mice were dosed prenatally or postnatally before puberty support that BPA exposure disrupts normal meiosis progression, with effects observed during meiosis I and meiosis II. And I know that we'll hear about another study that examined these endpoints in monkeys in a little while, which I think also adds to that.

The next endpoint I'm just going to briefly talk about is oocyte nest breakdown and assembly of two of the recent studies that I already mentioned regarding meiosis, the Chao et al. from 2012 and Zhang et al. from 2012 examined oocyte nest breakdown and follicle assembly and follicle recruitment.

So in the Chao et al. study they found that there was dose-dependently decreased primordial follicle numbers and increased follicles at later stages of development without an effect on the total follicle numbers at postnatal day 15 and 21 and that is with the two different dosing paradigms that I talked about early, either starting on postnatal day 7 or 5 respectively, and ending the day before the ovaries were collected.

So the results are consistent with accelerated recruitment of primordial follicles into the growing pool following dosing with BPA during this window. They also

observed increased expression of mRNA and protein of estrogen receptor-alpha at both time points, but not beta.

Zhang et al. reported -- they looked at earlier time points at oocyte and follicle numbers and they reported increased percentages of oocytes in cysts, that is not packaged into follicles, and decreased percentages of oocytes in primordial follicles at postnatal day 3. And this is during the time window in rodents when oocyte cysts break down and primordial follicles are formed in the 80 microgram per kilogram dose group, but no differences at subsequent days at postnatal day 7 -- 5 and 7, suggesting that the BPA treated catch up in that regard.

They also reported an increased number of total oocytes per section at postnatal day 3 and fewer total oocytes per section at postnatal day 7 in that group.

In a supportive paper from 2014, Zhang et al. treated cultured neonatal postnatal day 1 ovaries for 3 days with 0, 10, and 100 micromolar bis -- BPA. And they observed increased percentages of naked oocytes or -- oocytes and cysts, and decreased percentages of primordial follicles at both concentrations. So those in vitro data support what they had observed in the in vivo study, consistent with delayed oocyte cyst breakdown caused by perinatal exposure to BPA.

1 Because both of these studies had some flaws that
2 I mentioned earlier, I think that these endpoints need to
3 be further examined in additional studies, but I think
4 these results are certainly suggestive of effects of
5 bisphenol A on cyst breakdown and primordial follicle
6 recruitment.
7 So next I'm going to talk about developmental
8 effects exposures during the early life stages on the
9 ovaries, oviduct and the uterus in adulthood. So in
10 addition to a number of studies that reviewed the
11 uterotrophic effects of treatment with BPA in adult
12 rodents by various routes and doses that were summarized
13 in the DART document from 2009, as well as some other
14 endpoints uterine endpoints, multiple studies in mice and
15 rats have also examined the effects of early life exposure
16 to BPA on the ovaries, the oviduct, and the uterus in
17 adulthood. And cystic ovaries and uterine lesions were
18 observed in some of these studies in several strains of
19 rats and mice. And I'll mention some of those now.
20 So two studies by Newbold et al. from 2007 and
21 2009 examined the effects of early postnatal, so the
22 period of oocyte nest breakdown, and prenatal, gestational
23 day 9 to 16 the period when the gonads differentiate and
24 meiosis begins in the female. They both -- both of these
25 studies do subcutaneous injections of 0. And then in the

1 study that examined only prenatal dosing, they used 0.1
2 and 1 microgram per kilogram per day. Both studies then
3 used higher doses of 10, 100, and 1000 micrograms per
4 kilogram per day on ovarian, oviductal and uterine
5 histology at 16 to 18 months of age. So they age the mice
6 for about a year and a half before examining
7 histologically the tissues. And this was in CD-1 mice,
8 which is a strain that this group has published
9 extensively on the effects of DES, an estrogenic drug.
10 They observed increased prevalences of uterine,
11 ovarian, oviductal lesions with both of the dosing
12 windows. Prenatal dosing caused significantly increased
13 benign ovarian cysts and benign cystic endometrial
14 hyperplasia in the 100 microgram per kilogram BPA group
15 only, but nonsignificant increases in all other BPA
16 groups.
17 Also, the BPA groups had non-significantly
18 increased paraovarian Wolffian duct remnant cysts;
19 progressive proliferation of the oviduct, which was not
20 observed in the controls at all; uterine -- benign uterine
21 adenomyosis; Wolffian duct remnants in the uterine wall,
22 which was also not observed in the controls; a neoplastic
23 precursor to sarcoma, stromal polyps; leiomyoma, which was
24 not observed in the control and is a neoplastic lesion;
25 and atypical hyperplasia, which was also not observed in

the control, another premalignant lesion.

With the prenatal dosing window, the total incidence of ovarian and reproductive tract lesions increased in the BPA groups, with the highest incidence of 36 percent observed in the 0.1 microgram per kilogram group, followed by the 1 microgram per kilogram group. And both of those were significantly different from the control group.

For individual lesions in this prenatal dosing study, only the ovarian cysts in the 1 microgram per kilogram bisphenol A significantly differed from the control, but the pattern overall suggested the BPA effects. Ovarian cyst adenomas, tumors of the ovaries, were found in 10, 100, 1000 microgram per kilogram groups. And progressive proliferative lesions of the oviduct were seen in all BPA groups, with none in the controls.

Uterine Wolffian duct remnants were observed at 1, 10, and 1000 microgram per kilogram BPA groups only, none in the controls. Atypical uterine hyperplasia was observed in the 0.1, 1, and 1000 microgram per kilogram groups, none in the controls. And stromal polyps and stromal sarcoma -- or stromal sarcoma were observed in the 0, 1, 10 and 100 microgram per kilogram groups.

Several other studies also used subcutaneous dosing with similar microgram per kilogram dose ranges and

also higher doses in mice or rats and reported similar abnormalities.

So cystic ovaries and decreased numbers of corpora lutea were found by Adewale et al. in 2009 in Long Evans rats, and Fernandez et al., in 2010 in Sprague-Dawley rats after gestational dosing with BPA, and in BALB-C mice by Signorile et al. in 2010 after gestational and neonatal dosing with BPA.

The Fernandez et al. study additionally found lack of ovulated oocytes or offspring in the 50 milligram per kilogram per day group and decreased offspring production in the 5 milligram per kilogram per day group.

And the Signorile et al. study in addition found -- reported a trend for increased uterine precursor lesions adenomatous hyperplasia with cystic endometrial hyperplasia and atypical hyperplasia in the BPA groups at 3 months of age. They also found a significantly increased incidence of endometrial glands and endometrial stroma in the adipose tissue surrounding the pelvic organs in the BPA-treated animals. And those lesions stained positive for estrogen receptor-alpha and HOXA10, which are endometrial markers.

So the final set of endpoints and studies that I'll discuss have to do with mammary gland and early life exposure effects on the mammary gland. There are two

1 studies that were included in the 2009 DART review that
2 reported increase in terminal end buds in mice treated
3 subcutaneously and rats treated by gavage during gestation
4 with BPA.
5 The first study, Munoz-de-Toro et al. in 2006.
6 In that study, they dosed pregnant CD-1 mice with 0, 25,
7 and 250 nanogram per kilogram bisphenol A by subcutaneous
8 minipump from gestational day 9 to postnatal day 4 for 14
9 days. And then pups were culled and they were killed at
10 postnatal day 20, 30 and four months on proestrus for the
11 latter two time points. Some perinatally exposed animals
12 were additionally were ovariectomized at postnatal day 25
13 and treated with estradiol, 0.5 microgram per kilogram for
14 10 days to examine the effect of bisphenol A on the
15 mammary response to estradiol.
16 So they observed no effects on terminal end buds
17 at postnatal day 20, but they did observe a dose-dependent
18 increased number of terminal end buds and terminal end bud
19 area per ductal area at postnatal day 30. There were no
20 difference in the estradiol levels at first estrous or in
21 estrogen receptor-alpha positive epithelial cells at
22 postnatal day 90 -- 30.
23 There was an increased mammary response in terms
24 of the number of terminal end buds, area of terminal end
25 buds, the number of terminal end buds per ductal area, and

terminal end buds area per ductal area in response to estradiol in perinatally exposed BPA animals compared to controls.

They found decreased apoptosis measured by TUNEL in epithelial cells at both BPA doses on postnatal day 30, and increased progesterone receptor positive epithelial cells at that time point. And they also looked at the four month time point and there they found an increased number of side branches per ductal length at the 25 microgram per -- I'm sorry, nanogram per kilogram dose and nonsignificantly at 250 dose.

A second study by Moral et al. in 2008 dosed pregnant Charles River Sprague-Dawley -- or CD Sprague-Dawley rats, 8 weeks old by gavage with 0, 25 or 250 microgram per kilogram in sesame oil from gestational day 10 to 21. And offspring were euthanized at 21, 35, 50 and 100 days on the estrous stage for the last three.

Terminal end buds in this study were increased in the 250 relative to the 25 microgram per kilogram dose at 21 days, but not at the later time points. The number of terminal ducts increased dose-dependently at both 21 days and 100 days. And microarray analysis showed upregulation of differentiation genes at postnatal day 50 and downregulation of those same differentiation genes at post -- many of the same post -- genes at postnatal day 100 at

the 250 microgram per kilogram dose. There was also a cluster of immune genes that was upregulated at different time points in both doses.

So the question then arises do these mammary gland developmental changes have consequences later in life? And I'm going to just briefly talk about some of the evidence that mammary tumors develop in these animals -- in animals exposed to bisphenol A gestationally.

So Newbold et al. in the study that I already discussed, in which animals were exposed gestationally to bisphenol A, found two grossly evident mammary tumors, even though they were not screening mammary glands histologically in that study. And they were both adenocarcinomas.

And as described in the 2009 DART document, Murray et al. in 2007 reported that BPA dosing via subcutaneous minipumps during the gestational -- gestational day 9 to birth caused preneoplastic mammary lesions, ductal hyperplasias, as well as ductal carcinoma in situ in Wistar-Furth rats at postnatal day 90 and 95.

Importantly, in a more recent study by the same group - which I'm not sure was in the materials we got, but might have been, there were so many - is Acevedo et al., from Environmental Health Perspectives from 2013. In

that study, they treated Taconic Sprague-Dawley rats with 0, 0.25, 2.5, 25, and 250 microgram per kilogram BPA by minipump subcutaneously from gestational day 9 for 14 days or 28 days.

And they observed atypical ductal hyperplasia in all but one group and ductal carcinoma in situ in one group at postnatal day 50, and malignant adenocarcinomas were found at postnatal day 90, 140, and/or 200 in all the groups, either from the gestational day only dosing or gestational plus lactational, as well as benign lobular alveolar hyperplasia in the 250 gestational dosing only and the 25 gestational plus lactional dosing.

So although none of the individual groups was significantly different from control incidence, none of the controls had any of these lesions and they -- the combined N for the three time points was 23-35 per dose group with a total incidence of 1 to 2 per group. So this is important because it shows mammary carcinoma development following perinatal exposure at environmentally relevant doses of BPA.

So overall, I think together the weight of the evidence supports that gestational exposure to environmentally relevant doses of BPA alters mammary gland development in mice and rats and causes preneoplastic and neoplastic lesions in rats.

And in addition, just to summarize, what I've been talking about, I think the studies that have been published since the DART review in 2009 regarding early life exposures that the weight of the evidence is sufficient to conclude that early life exposure to bisphenol A has -- causes meiosis errors in females and lesions of the ovaries, oviduct, and uterus, as well as alterations in mammary gland development and alterations in sex differentiation.

Thank you.

CHAIRPERSON GOLD: Thank you very much, Dr. Luderer. Before we go to Dr. Pessah, any questions for Dr. Luderer?

Okay. Dr. Pessah.

COMMITTEE MEMBER PESSAH: Thank you. That was a very extensive review of the developmental literature. I took a slightly different approach. I basically asked the question is what's the typical range of concentrations or levels in humans of population based measurements?

And the best values I can come up with is somewhere between 1 and 20 nanomolar, which translates into about less than 50 nanograms per milliliter of either serum or plasma. That was from Welshons et al. It's an old study but highly cited in Endocrinology in 2006.

There was also some evidence that during

gestation that there's accumulation of BPA in gestational tissues, and that could be as much as five-fold, which results in a level that could be around 100 nanomolar, which is a little less than 250 nanograms per milliliter.

And so in reviewing the vast literature in animal studies, I do believe the weight of evidence suggests that BPA exposures during gestation have the potential to affect at least two early stages of oogenesis. The onset and rate of meiosis in fetal ovaries during the primordial to primary to secondary follicle transition and the rate and integrity of germ cell nest breakdown and follicle development, this apparently occurs without causing gross chromosomal damage, such as aneuploidy.

However, when I read the literature, I found the most recent data, and perhaps the most compelling data, comes from recent studies that indicate relatively low levels of exposure in vivo, influenced subtle changes in epigenetic dynamics, and influenced differentially methylated DNA regions, or DMRs.

Such modifications at tissue-specific DMRs appear to be complex and highly dependent on the timing and level of BPA exposures. The BPA-induced epigenetic changes in maternally imprinted genes are especially a concern, since they are likely to have an impact on gene expression patterns, not only in the gestationally exposed F1, but

are likely to endure and be transmitted transgenerationally.

The impact of such epigenetic modifications and how they influence neurodevelopmental outcomes in health over the life span are just beginning to be understood. And that's where the literature really is very, very young.

That said, I did review some of the papers that Dr. Luderer presented, and it led me to conclude that BPA has the potential of elucidating reproductive and developmental changes. The results from such study are greatly divergent in their findings, and suffer from the lack of defined dose-response relationship.

These are probably hampered by the spatial and temporal complexity of oocyte follicle development, that is that because the ovaries and follicles are small, especially from small experimental animals, that defining different regions is difficult and, in fact, the average may not really represent what's actually happening within the follicle developmental transition.

The other issues that I had with most of the literature was that biological plausibility seems to be lacking, in terms of target engagement. That is, is it plausible that estrogen receptors are altered sufficiently to cause observable -- the observable outcomes that were

being measured in the study?

So to highlight some of these, I focused in on a handful of studies that try to measure dose-response or concentration-effect relationships, both in vivo and in vitro, and studies that seemed to measure the same outcomes in similar species.

So the first study is Lawson in 2011, used time-mated C57, and had a single exposure level of 20 micrograms per kilogram of BPA. This was administered orally in corn oil. And the dosing began at post-conception day 11. And then samples were taken at post-conception day 12 through 14.5.

They did an excellent baseline analysis of meiotic genes that were expressed during that time period, and showed that there were 16 of 18 important meiotic genes, that is genes that regulate meiosis in oocytes or early gametes and germ cell development. Some of these were increased more than two-fold, some essentially about five-fold. So this was in the wild-type situation, in other words untreated animals.

BPA exposure seemed to increase a handful of these, including a particular gene that's the stimulated by retinoic acid 8 homolog, or Stra8, that several studies measured, and found it to be increased at least two-fold or about two-fold. I found that compelling, but of the 16

of the 18 that were seen to increase in that developmental window in untreated animals, only three BPAs were differentially expressed during that same developmental time period. The trends were the same though with BPA treatment and without. The first changes were evident within 24 hours of exposure, but most extensive changes were in that critical period right around 14.5 post-conception.

There was also a downregulation of mitotic cell cycle genes. So this indicated that fetal BPA could in fact not only influence meiosis, and genes that regulate meiosis, but also could include influence the expansion of primordial germ cell populations.

Zhang et al., and that's X. Zhang in 2012, changed the exposure period by doing -- initiating exposure at post-conception day 0.5, as opposed to the Lawson study, which began at day 11, post-conception. And they actually found a downregulation of about 70 percent in expression of meiotic genes, such as Stra8 and Dazl. Both of these were shown to be significantly downregulated, as opposed to the Lawson paper, which showed an upregulation.

Now, this may be due to the difference in timing of exposure when it commenced. They also saw a rather large change downregulation of about 80 percent in

transcripts for a homeobox gene that regulates oogenesis. That's homeobox or Nobox as it's called. And this occurred both in females and males. There was no sex difference.

This was all done in CD-1 mice. They did look at DNA methyl imprinting genes. And these, in particular, was IGF2R, peg 3, and H19. There was a general decrease at the highest dose of 80 and 160 micrograms per kilogram per day BPA. Again, these were administered orally in DMSO.

And in terms of estrogen receptors, they saw a increase of about two-fold in the highest dose, 160 micrograms per kilogram per day. It was no change in the ER beta.

So neonatal exposure to BPA seems to differentially inhibit or enhance methylation of imprinting genes and meiotic genes during oogenesis, but the effects appear to be variable and may be somewhat stochastic, because the dose response really doesn't suggest that there is a linear or logarithmic dose response relationship.

In a follow-up study I think by the same group, although the lead author is Zhang H.Q., CD-1, same strain of mice, were exposed to 20, 40, 80 micrograms per kilogram per day orally in DMSO at pre- -- sorry at

conception day 12.5 to 18.5. So now they've shifted to later exposure initiation. Ovaries from pups in both treated and control groups were examined at postnatal day 3, 5 and 7. They HRP staining, and they found that the number of oocytes per cyst were increased 3.5-fold, but did only at the highest dose of 80 micrograms per kilogram BPA.

They also showed that at the highest dose, there was only an effect at the primordial follicles not the primary, secondary follicle stages, which is surprising since you'd think that that would carry over.

There was a very modest decrease, unlike their first study in 2011, which showed almost an 80 percent decrease in transcripts for meiotic gene regulators. They showed a very modest but huge error, but still statistically significant decrease, and no change in other mitotic genes tested. And so this was a small difference in Stra8, but none of the others seem to show a difference. BPA significantly activated ER alpha expression, and no effect was observed on ER beta. This is in contrast to an earlier study by Susiarjo in 2007 that showed that BPA had early meiotic effects.

So collectively, again these studies seem to indicate that, although variable, BPA can inhibit primordial follicle assembly by regulating some meiotic

genes. But again, these observations do vary from study to study in rodents.

In vitro. In Enriquez, two papers, one in 2011 and one in 2012 used, what I consider, very high levels of BPA, 1 to 30 micromolar, and showed that increased oocyte degeneration by impairing meiotic progression in cultured human oocytes. The data I considered is very weak and the data -- the figures actually have typos in them. And the changes are very, very small, in other words, less than 10 percent changes with significant errors associated with those changes. Nevertheless, they report statistical significance.

Trapphoff in 2013 actually did a study on C57 oocyte follicle cultures, where they used very low concentrations of BPA, 3 to -- or 300 nanomolar as opposed to the supramicromolar that Enriquez used. And they looked at methylation of differentially methylated regions of DNA, which is very important, especially those that are known to be maternally imprinting regions of DNA. They showed a non-monotonic dose response curve, where the effects were significant at 3 nanomolar, but not at 300 nanomolar. And so there seems to be a non-monotonic dose response at least at the two concentrations, which are 100-fold apart.

They also showed that paternally imprinted genes,

such as 819, in mouse germinal cells were altered. Trimethylation of histones H3K9 and acetylation of histone H4K12, these are important in early germ cell development, and the distance between centromere of sister chromatids in metaphase II were also impacted.

So the conclusion is that these very low levels, 3 nanomolar but not 300 nanomolar caused slightly accelerated follicle development, which is statistically significant, and also statistically significant methylation errors in differentially methylated regions of DNA. This is particularly significant, because some of these were seen to occur at maternally imprinted genes which could have ramifications downstream.

So in terms of germ cell breakdown, there's several in vivo studies. I actually looked at a couple of them. Veiga-Lopez in 2013 had a, I thought, a very well done experimental design. They exposed in the prenatal period to BPA at 0.5 mg/kg per day subcutaneously. This was done in Hughes. They actually -- this is one of the few studies where I actually saw blood levels reported as part of the experimental outcome. And they reported mean levels of about 2.6 nanograms per milliliter, which is in the range, and this was unconjugated free BPA, in umbilical arterial samples. And this approaches the median levels of BPA measured in maternal circulation. So

I found this study rather compelling.

They reported that expression of stereogenic -- steroidogenic enzymes and steroid gonadotropin receptors and key ovarian regulators and micro RNA biogenesis, using RTPCR and nested design RTCPR, that there was an age-dependent effect in most steroidogenic enzymes that regulate ovarian development.

But BPA -- so this is what they saw over time in untreated animals, BPA-treated animals seemed to differentially alter a couple of these genes, including the steroidal regulatory gene or metabolic gene SIP 19 that was altered upregulated by about two-fold and SDRA5A1, which was downregulated about 1.5-fold.

But this was only at gestational day 90 and not at gestation -- I'm sorry at gestational day 60 -- 65, but not gestational day 90. And in terms of steroid receptors, none were altered by BPA across this time period.

So in terms of microRNAs and their expression, they were altered by this prenatal BPA exposure. Forty-five of these were downregulated at least 1.5-fold at day 65 and by day 90, 11 were downregulated. These included several genes that -- or microRNAs that regulate oocyte development.

So the results from this study suggest that

exposure to BPA at environmentally relevant dose and at doses that are relevant in this circulation alters fetal ovarian steroidogenic gene expression and microRNA -- patterns of microRNA expression that are relevant to gonadal differentiation, folliculogenesis and insulin homeostasis.

A paper by Rivera et al. followed up with again neonatal exposure to use at 50 micrograms per kilogram per day. This considered the EPA safe dose. And this was exposure preceded early neonatally between postnatal day 1 and 14 -- on postnatal day 1 and 14 daily.

The ovaries were analyzed on postnatal day 1, 5, 10, and 30. It was a robust study with three individuals per time point and three samples per time point. And they used this design to describe the spatial and temporal pattern of expression of estrogen receptors, alpha and beta, androgen receptor at using immunohistochemistry.

Hormonal levels were obtained from blood serum. And the key findings were that BPA at the 50 microgram per kilogram per day level accelerates germ cell nest breakdown at the antral, about 10 to 15 percent, change in primordial to transitional to primary, that is the primordial to primary follicle stages, but not the preantral stages of follicle transition.

It's unclear how this relates to a very, very

large change in the expression of P57, which is a cyclin-dependent kinase inhibitor. It actually puts the break on cell division, but they found a very, very marked increase in P27 expression.

Rodriguez in 2010 did a rat study at 0.05 and 20 milligrams per kilogram administered subcutaneously every 48 hours on postnatal day 1, 3, 5 and 7. They saw about a two-fold decrease in primordial follicle expression, and an increase in follicle recruitment. There was no change in multi-ovarian follicles with BPA exposure.

BPA, at this level, produced a very, very marked rate of a four-fold increase in P27 expression in primordial, primary, and transitional follicles consistent with the lamb study of Rivera. So the P27 result seems to be a consistent finding across species.

So P27 is a CDK1B expression, which regular -- expresser, which regulates cell cycle programming at G1. So it was a rather important regulator of cell cycle.

A recent study by Li et al. in 2014 used high BPA exposures, a little later during postnatal development, essentially at pre-puberty, between postnatal days 21 and 27. They administered BPA intraperitoneal at 10, 40 and 160 milligrams per kilogram per day.

This decreased the number of all follicle types and increased atretic follicles in the rat, and suggested

that this could lead to premature reproductive senescence, but this, of course, needs confirmation since they didn't measure that. A weakness in this study is that BPA exposure groups were basically IP at rather high levels of BPA.

The most dramatic effects were seen at 160 milligrams per kilogram per day. They saw some decreases in oocyte specific histones such as H1FOO, but this was not dose dependent. It only occurred at the highest level of exposure. There was no change in estradiol. And the dose response in progesterone was seen, but not -- there was no effect on estradiol.

Let's see here. In vitro studies to -- to look at this type of effects of BPA, this was essentially the Peretz review concluded that in vitro exposures strengthen the weight of evidence that BPA effects onset of meiosis. But if you look at their study in 2011 and 2012, the in vitro studies used 4 and 440 micromolar of BPA in FVB mouse ovaries that were harvested on postnatal day 32.

Antral follicles were mechanically isolated from these ovaries. BPA at 440 micromolar decreased antral follicle growth throughout the 120-hour culture period and decreased estradiol and estrone, testosterone, androstenedione, DHEA and progesterone levels that were produced by these follicles. But again, this was at 440

micromolar.

At 50 micromolar, they saw upregulated expression of cell cycle regulators and the pro-atretic and anti-atretic factors BAX and BCL2 associated protein. That's what BAX is. TRP53, which is a tumor promoter protein, and BCL2.

Unfortunately, there was no dose response relationship and a non-monotonic dose response relationship was shown for expression of ER alpha and beta, where 5 micromolar BPA caused a maximal response, whereas 0.5 micromolar and 50 micromolar had no effect on either side of that. Not sure what to make of that.

So there's several studies that look at steroidogenesis in females in vivo. Three experimental studies have shown that BPA exposure alters ovarian steroidogenesis in the perinatal period. That's Xi et al. in 2011, the postnatal period, that's Fernandez, 2010, and Tan in 2013. And basically, that these studies seem to have variable results of which steroids are altered and which steroidal enzymes are altered and how they're altered.

So, for example, Fernandez in an EHP paper in 2010 used the SD rats that were exposed at postnatal day 1 through postnatal day 10 orally in castor oil at levels of 0.62 to 62 milligrams per kilogram. They saw an increase

in estradiol and testosterone and a decrease in progesterone. But again, what you see is a step function. No effect at the low dose, and then a saturating effect at the two higher doses. In terms of the progesterone, there seemed to be more of a dose-response relationship there.

Now, it should be noted that these changes in estradiol the increases are about 30 percent over baseline, and testosterone is about less than two percent -- sorry, two-fold increase. The estradiol result does not replicate a previous study by Berger at al. in 2008 and does not replicate a more recent study by Lee et al. in 2013. The Lee et al. in 2013 is an Environmental Health Perspectives paper where adult rats were exposed at 1 or 10 micrograms per kilogram per day orally. This resulted in about a three-fold reduction in estradiol, not an increase. So that seems to be at odds, and a two-fold reduction in testosterone.

They also saw a two-fold increase in apoptotic markers, such caspase-3, steroidogenic proteins, StAR or StAR for short, and P450 aromatase, which is essentially CYP 19. So these appear to be targeted by BPA.

Xi in 2010 reported that postnatal BPA exposure alone actually did not affect serum hormone levels in mice. In four other studies using rats, mice, lambs, at gestational or gestational plus neonatal exposure to BPA

at lower doses, less than 20 milligrams per kilogram had no effect on steroidogenesis. And this includes a study by -- a -- Kobayashi in 2012, Mendoza-Rodriguez in 2011, Rivera in 2011 and Varayoud in 2011. So the results of in vitro studies on the effect of BPA and steroidogenesis are somewhat variable.

I'm going to fast forward to recent studies that look at mechanisms of BPA toxicity. There are a couple of compelling papers that have come out in 2013, in particular Tang et al., which used trying to get at the idea of how do the changes that were described this morning, how are they manifest. Are they manifest by direct interactions with steroidal receptors or do they change enzyme profiles that regulate steroid metabolism?

So they used Hexcel that's stably express individual nuclear receptor ligand binding domains. These were linked to a reporter -- betagal, and they examined high quantitative, high throughput screening of a format that is implemented in Tox21 at the NIH.

Two receptors, estrogen receptor alpha and androgen receptor seem to be directly affected by BPA. And these are affected in opposite directions, supporting the idea that there may be a differential regulation by which BPA causes its sex-specific effects. To confirm these observations of BPA on the estrogen receptor and the

1 androgen receptor, they performed transient transfection
2 experiments with full length receptors and look for their
3 corresponding response elements linked to luciferase
4 reporter.
5 So what they showed was that, in fact, BPA and
6 congeners of BPA, such as BPAF, act directly on
7 androgen -- estrogen receptor-alpha as an agonist with a
8 half maximal effective concentration in EC50, of about 200
9 nanomolar. Now, that, I think, is significant, because
10 this is considered a high affinity effect. But it should
11 be noticed that it's greater than 4 log units lower than
12 estrogen itself, estradiol itself.
13 As an AR receptor -- androgen receptor
14 antagonist, BPA is an incomplete antagonist that only
15 partially inhibits the receptor, even at the highest doses
16 that they use, the highest concentration they use, and
17 they can't really calculate an IC50, which probably is in
18 the neighborhood of greater than 100 micromolar, if one
19 had to estimate.
20 Again, speaking to behavioral effects on BPA
21 exposure during gestation, there's been a study recently
22 published -- and, I'm sorry, I didn't have the -- oh
23 Susiarjo 2013 showed that BPA alters expression of key
24 genes in the placenta. The majority of the affected genes
25 were also expressed abnormally in the placenta and other

parts of -- and other tissues. And DNA methylation studies showed that BPA significantly altered methylation levels of differentially methylated regions, DMRs, which I spoke to at the beginning as being a compelling evidence that there could be long-term effects that aren't seen through immunohistochemistry and so forth.

These include imprinting genes such as SNRPN and IGF2, which replicates a previous study. And there's also some genome-wide changes in methylation in the placenta, but actually those global changes are not seen in the embryo.

So there seems to be a critical window of susceptibility in terms of when the animal studies are initiating and terminating BPA exposure. These seem to be somewhat variable from study to study, but all studies seem to show biological effects of BPA exposure during the perinatal period, especially compelling to me were the effects in early meiosis, which seem to be reproduced across species and across developmental windows exactly how those changes occur seem to be dependent on timing and species.

I think the -- again to conclude the evidence of changes in methylation of DNA differentially methylated regions of DNA, especially maternal imprinting regions seem to be compelling and deserve more work.

And I think I'll stop there.

CHAIRPERSON GOLD: Thank you very much, Dr. Pessah. Anyone have questions for Dr. Pessah?

Okay. I think we have time to start the next section, which is on non-human primates and other mammals, which Dr. Kim, you're going to start with. Thank you.

COMMITTEE MEMBER AUYEUNG-KIM: Yes. Thank you very much.

So the wealth of information for the non-human primate, as well as other species, which I covered the sheep that Dr. Pessah spoke about as well, many of the findings that were observed in the mice or the rodents were also tested in the non-human primate and sheep. And so the -- most of the studies primarily focused on early oogenesis and ovarian follicle formation and steroidogenesis.

In the non-human primate, there was a set of studies conducted by a group that used female Rhesus monkeys. And what they -- in this set of studies, I commend the group for utilizing these set of monkeys for attaining a wealth of information to help the progression of BPA -- understanding the mechanism of action of BPA.

However, this leads to a limitation in that all existing NHP data were generated using the same cohort of animals, so that should there be certain predisposition

unrelated to the BPA administration, it could bias the data in all the studies. In these studies, the Rhesus monkeys were treated -- they were broken up into essentially two cohorts, of which they were -- those two cohorts were subdivided into two different time periods of -- or -- so the -- the non-human -- the Rhesus monkeys were treated either in the early treatment group, which is GD 50 to GD 100, which is the second trimester where germ cell differentiation and meiotic entry occurs or they were treated during the late treatment, GD 100 to term, which is the third trimester when follicle formation takes place.

These two groups were subdivided where there was a cohort that had a single daily dietary dose of 400 micrograms per kilograms per day. Typically, it was five to six treated BPA-treated animals and control animals. And the other group was a continuous exposed animals, which were dosed with a intradermally placed place silastic capsule and this cohort of animals were six treated and two controls.

The single and continuous exposure animals were connected during different breeding seasons. Therefore some results differ between the two groups, potentially due to the different levels of exposures to the phytoestrogens that could be related -- that could be in

the feed or due to the limited number of control animals in the control group for the continuous exposure group.

The benefits of this is that there was one PK study that was conducted and PK data was made available in which -- and this was presented in the Taylor et al. paper in 2011, where it found that the average exposure for the non-human primate was 0.52 nanograms per ml, and then mice it's 0.5 nanograms -- was approximately 0.5 nanograms per ml, each with a lower limit of connotation of 0.2 nanograms per ml. And in human, based on previous data, generally the exposure was 2 nanograms per ml of the unconjugated BPA.

So this first study that I want to talk about is Hunt, which this is similar to what was discussed by Dr. Luderer, as well as Dr. Pessah, is that BPA exposure induces changes in meiotic chromosome behavior. And this disrupted the synapsis and recombination that occurs between the homologous chromosomes at the onset of meiosis. And this is consistent with observations that were reported in the mice.

BPA also disrupts the follicle formation, in that there was an increased number of multi-oocyte follicles in the antral and secondary follicles at birth. And they were observed in both. So the multi-oocyte follicles were observed in the single daily dose cohort, but not in the

continuous exposure cohort.

And then there was -- and then in the reverse in the continuous exposure cohort, there was an increase incidence of unenclosed oocytes, but not -- but that was not observed in the single daily dose cohort. And so -- but the strength of these studies that -- the findings that were observed in these studies were similar in -- were also observed in the rat and mice, lamb that Dr. Pessah spoke about, and then also in in vitro studies.

The next study is Dr. -- is the Calhoun study, which this only looked at the single daily dose cohort, where at GD 165 there was significant differences in gene expression compared to controls. The genes that were critical for reproductive organ development in are adult functions was HOXA13, WNT4, and WNT5A.

So although there were changes in these genes expressions, there were no effect on the histology or cell expression, the proliferation marker -- there was no effect on histology, and there was also no changes in the proliferation cell markers KI67, ER alpha, and PR compared to the control. Oh, and in this study, they used microarray histology and IHC.

So the strength of this study is that the BPA exposure does not significantly affect the fetal uterus development as evidenced by morphologic and steroid

hormone assessments. The third study is Tharp, 2012, where they looked at mammary glands in the neonates exposed to BPA in utero. There were more developed than the controls for -- they were more developed than controls for terminal buds, the terminal ends, the branching point, the bifurcation ends, and total mammary area, including the ductile area and the number of ducts. Some were not statistically significant however.

And there was no difference in the expression of the ER alpha and ER beta, compared to control. The strength of the study is that the mammary gland effects have been observed in mice, rats, and monkeys -- and now monkeys. And it could suggest that BPA could have developmental effects on the mammary gland, but the studies do not clearly show breast cancer risk or effect on the function of the mammary on its own stands.

There was one additional study by Dr. Aldad that was conducted in African green monkeys. And the low dose -- in this study, African green monkeys, the agent husbandry information was not provided. There was a single dose -- single dose administered by silastic capsule continuing a mini-pump. And it did not indicate when the treatment started after oophorectomy.

But in this study, the low dose of BPA did not affect the progesterone receptor expression, but the BPA

1 dampened the glandular and stromal progesterone receptor
2 expression in response to estradiol. In combination with
3 estradiol, the BPA diminished the ETU-induce endometrium P
4 receptor -- the PR receptor. And so this again -- this
5 also -- this -- sorry. So this shows that in this study
6 the BPA is shown to affect steroidogenesis.
7 And then there was several sheep studies that
8 were conducted, of which Dr. Pessah did touch on several
9 of them by Salloum 2013 and Veiga-Lopez. So I won't
10 discuss those because similar to what he presented are --
11 is -- are the conclusions that I reached as well.
12 And so there was a paper for Evans that the
13 conclusions of the study was that the exposure prepubertal
14 female lambs were exposed to BPA. And this was a single
15 dose of 3.5 mg/kg per day. That was administered
16 biweekly, intramuscular for 7 weeks. And it showed that
17 it can suppress -- BPA can suppress gonadotropin secretion
18 and -- as demonstrated by the LH, pulse, and amplitude and
19 frequency, but there is no effect on the LHRFSH profile
20 compared to controls.
21 And in another study, the Salloum study, prenatal
22 exposure to -- and this is also in lambs that were exposed
23 during gestation day 30 to 90, the BPA -- there was an N
24 of 8. They were exposed to a single dose of 5 mg/kg of
25 BPA and it reduced -- and in this study it showed that

there was reduced sensitivity to estradiol and progesterone negative feedback. There was increase in pituitary responsiveness to gonadotropin releasing hormones. And this dampens the LH surge response to estradiols positive feedback challenge. So similar to the Evans paper that there was a decrease in the LH surge.

So as far as these studies are concerned, I think standing on its own for the non-human primate studies, as well as the sheep studies, that on their own there are limitations to the conclusions of the study, because in most studies there was only a single dose level that was administered. And whether those doses were -- although the exposure may be the same, the route of exposure that was administered was subcutaneous or through a mini-pump.

And so whether it's relevant to human exposure remains to be seen. However, considering the weight of evidence presented with the rodent studies, as well as in vitro studies that there -- this -- this could show that there is cause for concern, whether BPA is a reproductive toxicant.

CHAIRPERSON GOLD: Thank you.

Any questions?

Dr. Plopper, are you ready?

COMMITTEE MEMBER PLOPPER: Well, thank you. I think that the last three speakers actually covered all of

the studies that I was assigned to review.

(Laughter.)

COMMITTEE MEMBER PLOPPER: So that will bring everybody to lunch.

(Laughter.)

COMMITTEE MEMBER PLOPPER: But I did -- I do want to say one thing and that is I took a slightly different approach, because I was concerned about the weakness of studies using large animals, specifically sheep and primates, and my first question was what is the exposure environment that the specific target organ or organ system is concerned with? How does it interact?

And the situation here, as far as from my experiences of teaching a lot of anatomy and physiology is that it's the circulatory -- it's essentially the arterial concentrations for reproductive organs, female reproductive organs, specifically the ovary, the oviduct, and the uterus, so that there is two issues to be addressed here.

One is, is the exposure appropriate based on what the levels are in the arterial system? And then secondly, what are the strategies used to put it there?

And, as has been emphasized already, the studies in non-human primates and sheep do not have dose responses, because that's just not practical.

So the issue is were the strategies they used, most of which may or may not have been relevant to human situations produce levels of circulating unconjugated BPA that are relevant to humans?

And the fact of the matter is that all of them did. And you heard that there were some significant changes here. And the ones that are critical, which by weight of evidence, would suggest that BPA is causing a problem in female reproduction, are, in fact, changes in meiosis, oocyte formation, and organization of the oviduct.

And I want to emphasize that if that seems like a concern, because in the primate studies, they used a single dose a day in a -- by fruit. And so if you follow the pharmacokinetics there, you see that the level goes very high for a very short period of time up in the upper end of the range identified in humans, and then it tails off over a 24-hour period.

The same types of changes were found there are found in all these other studies where there is a sufficient exposure pattern to keep the level high for the full 24 hours. Okay. That, to me, is a concern from my experience with this.

And the other is the silastic tubes that are used to do all these long-term continuous studies. I don't

1 believe that that is inappropriate, because that maintains
2 the levels that they identified in these animals is in the
3 same range that has been observed in people.
4 So we don't have a dose response here. We have a
5 zero and a level, but that level is within the range that
6 would be experienced by people. So they've already done a
7 nice job of explaining all the key things here.
8 It's not only genes get changed, but obviously
9 the oocyte formation, organization of follicles is
10 markedly changed in three different species at levels that
11 are experienced in people.
12 And the other thing that I would say -- I'll just
13 add one more thing about the Hunt study that is of concern
14 to me, is that a large percentage of those oocytes are not
15 associated with granular follicle cells at essentially a
16 newborn female. What that means is they're never going to
17 form.
18 In fact, from my pathology approach, I would
19 identify about 90 percent of those nuclei as being
20 pyknotic, which means they're about to die.
21 So I don't know -- I think there's a lot more
22 study to do, but I think that the weight of evidence
23 clearly shows, at least in terms of female reproduction,
24 that BPA, at levels experienced in the human population
25 does cause a problem.

I won't go into the details. They've already done all that. If you want to argue with me, I'll be glad to discuss it point by point.

Thank you.

CHAIRPERSON GOLD: Thank you very much. Any questions for Dr. Plopper?

I think it's time for a break.

Any questions before we take a break?

Questions. Questions.

Okay.

CHIEF COUNSEL MONAHAN CUMMINGS: This is Carol Monahan Cummings. Again, if we're going to take a break, I just want to remind the members that during breaks, you aren't allowed to talk amongst yourselves about the subject matter of the meeting. And my recommendation would be that you also not talk to third parties regarding that same information. If you do, then you just need to disclose the fact that you had a discussion with someone, and give the general content of that discussion, so that it's part of the public record.

Dr. Gold, did you have a certain amount of time you were thinking about?

CHAIRPERSON GOLD: I'm thinking it's a good time for a lunch break. And maybe does 12:30 sound reasonable to come back by?

Is that a problem for anybody?

Too short?

CHIEF COUNSEL MONAHAN CUMMINGS: We could mention also, if you didn't already, Dr. Zeise, that there is a cafeteria downstairs, such as it is, but it's quick. There's also a number of different restaurants in the very close vicinity where you can get sandwiches and things like that. If you need some direction, we can help you with that.

CHAIRPERSON GOLD: We'll all aim to be back at 12:30. Does that work for people?

Is it too soon? Should we make it 12:40?

CHAIRPERSON GOLD: 12:40. Okay. Thank you everyone. We'll see you after lunch.

(Off record: 11:53 AM)

(Thereupon a lunch break was taken.)

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

(On record: 12:45 PM)

CHAIRPERSON GOLD: Okay. I think I'll welcome everybody back. I do want to remind people if they -- the public, if they want to speak, they should get their blue card to Esther when she returns. She'll be back shortly.

Anybody from the staff have anything they want to -- we're good.

Okay. So I think we're ready to turn now to the human studies and Dr. Carmichael will start us off.

COMMITTEE MEMBER CARMICHAEL: So first I'm going to just make a few comments about my general approach to the review, and then highlight some of my major concerns with the literature in general. And then I'll summarize the findings by outcome. So my first step was to review each relevant study, and basically evaluate its potential validity.

So this is kind of taking a turn from the animal experimental literature. But in epidemiology, we can't assign -- typically, we can't assign the exposure of interest at random, and we rely on observational studies. So therefore, we have to pay careful attention to non-random factors that might affect the results or jeopardize the validity.

So for studies that I deemed to have potentially

good validity, the second step was basically to consider the consistency of results across studies for each outcome, and whether the evidence seemed to point toward an association.

So again, in epidemiology, we don't typically -- for the reasons for the first point, we don't typically rely on a single study for decision making. Rather, we look for consistency of results across various designs and populations. So I want to point out a few of my major concerns that were sort of a theme for this literature, or basically the major threats to validity of findings that come up most often.

So the first one I'll mention is temporality. So a lot of studies are cross-sectional -- of the epidemiologic studies are cross-sectional, which means that the exposure and the outcome were measured at the same point in time, so there's really no way to establish whether one -- which one came first. So what I focused on is a perspective design, meaning that the BPA levels were measured before the outcome occurred. And even then, timing still may not be optimal. It depends on the particular outcome and what we think the important window of vulnerability is.

Sample size is another concern. So, for example, at some point, if the sample is just so very small, then

the results may be imprecise and it's really hard to conclude -- hard to really even find a statistically significant association. So that's a limitation of some studies.

Selection bias is another important one. And this is just taken to be a general term referring to whether the selection of study subjects seems reasonable, and, in particular, whether the cases and the comparison group seem to be, for example, from the same underlying population or just, in general, whether it seems like they're a comparison between those two groups seems reasonable for the purpose of observing an association with an exposure.

Another concern is with confounding. So confounding is the issue where -- a confounder is a factor that is related to both the exposure and the outcome. And if that happens, then we're concerned that if we look at an association between the exposure and the outcome, whether it's attributable to that third factor. So, for example, if BPA and an outcome are both related to age or infant sex, then it's important that the analysis would adjust for that, that third -- that confounder, so that we know that the association is independent of that association. The association with BPA is independent of the associations with the confounder.

And then the last issue I'm going to mention is definitely an important one, and it is related to measurement error. So BPA has a very short half-life. And as such, a single BPA level, just one -- if there's just one measurement, it reflects very recent exposure, because the levels are highly variable within even just a short time frame. So I won't get into statistics today, but one statistic I want -- statistical test I want to mention in this context is the intraclass correlation coefficient.

And basically, this is a measure that kind of reflects that variability with -- and it estimates whether the variability with over -- across measurements made within one individual is greater than the variability between individuals. So basically it's calculated as the between-person variance divided by the sum of the between-person variance, plus the -- with the intra-individual variance.

So as a rule of thumb, this correlation coefficient, if it's greater than 0.8, it's considered that, you know, you have excellent reproducibility with repeated over time -- or with repeated measures. So if it's 0.4 to 0.8, it's considered fair to good. If it's less than 0.4, it's considered poor. So there have been a number of studies that have looked at the intraclass

correlation coefficient of BPA measures, and in particular, over short amounts of time. And they tend to be 0.1 to 0.2.

So that would be in the poor range. So that just -- basically, what that tells us is that really to get a good idea of BPA exposure in humans for -- to get a good idea of average exposure, it's likely that greater than one sample is preferable. Otherwise, because of all this -- because of this large variability, probably most of the time it's likely that the associations that we observe are attenuated or are weaker than we would expect.

So now, I'm going to summarize the findings by outcome. And again, I'll focus on the studies that, based on my review, seem to be of reasonable quality. For example, I am not reviewing the cross-sectional studies or not focusing on those and -- or other studies that I consider to have major or multiple major methodologic concerns.

So the first set of studies has to do with oocyte quantity and quality and fertilization. And these studies have been conducted among women undergoing IVF, or in vitro fertilization. And one set of studies was from UCSF clinic, and one set of studies was from a clinic at Massachusetts General Hospital.

So the UCSF studies were by Bloom and Fujimoto.

1 And just in summary, they found no association with oocyte
2 number or embryo quality, but they did find significantly
3 reduced fertilizations. And this was in about 30 to 40
4 woman for each study. And the BPA samples was one sample
5 collected around the time or shortly before oocyte
6 retrieval.
7 And then the Massachusetts General Hospital
8 studies, there are two by Ehrlich. And they did find an
9 association. They found about a 25 percent lower mean
10 number of oocytes, total number of oocytes, and number of
11 mature oocytes, and number that were normally fertilized
12 among women who had higher BPA levels. And they actually
13 had two measures of BPA, and they averaged them, one was
14 early in the woman's cycle and then one was the day of
15 oocyte retrieval.
16 And then there are a few outcomes I'll just
17 mention very briefly, because there was only one study per
18 outcome. So fecundability or time to pregnancy by Buck
19 Louis, there has been one study, and it did not find an
20 association. The odds ratio was 1.0. Spontaneous
21 abortion or miscarriage study by Lathi, found a
22 significantly increased risk for miscarriage with
23 increasing BPA. And the -- it was based on two BPA
24 measurements -- wait, yes -- for most women measured
25 shortly after conception.

1 There's been one study that I will mention that
2 was a prospective study looking at puberty by Wolff. It
3 basically found that looking at breast development,
4 looking at -- they measured BPA in girls when they were
5 six to eight years old and then looked at their breast
6 development a year later and did not see an association.
7 And there's been one study that I will mention on
8 endometriosis by Buck Louis. And basically that study
9 incorporated -- looked at two cohorts, so it's kind of two
10 studies in one. And there was a positive association
11 increased risk with increased BPA in one of the cohorts
12 but not the other.
13 But it -- the BPA measurements, there were single
14 measurements, and they were measured shortly before the
15 procedure -- the procedures that were done to assess
16 endometriosis.
17 So now I'm going to move on to the studies that
18 have to do with infant size and gestational age at
19 delivery, so pregnancy outcomes. So I'll start with birth
20 weight. There are three cohort studies that have been
21 done. And a study by Lee and a study by Philippat, both
22 found a significant positive association with birth
23 weight, that is higher levels of BPA were associated with
24 higher birth weight. And then Wolff and others did a
25 study that did not find an association with birth weight.

I will note that all of these -- yes, all three of them had only one BPA measurement, and it was measured in the third trimester. And I'm not clear on how much time there was between delivery and -- delivery and the measurement of BPA in some of these.

Another birth outcome that's been studied is gestational age. And there are two cohort studies that have looked at this outcome. Weinberger found that increased BPA was associated with a significantly shorter gestational age, and Cantonwine found there was -- that higher BPA was associated with significantly increased risk of pre-term delivery. And Weinberger had one BPA measurement and that was the last visit before delivery, so -- and then Cantonwine had a third trimester BPA measurement.

There's been one study that I will talk about that looked at term. It looked at growth retardation, so we refer to that as small for gestational age, and that was among babies who were born at term or at least 37 weeks of gestation. That's by Burstyn. And that study found an odds ratio of 1.0, which is basically a no association. And that study had one BPA measurement, which was taken mid-pregnancy.

And there are also a few studies that have looked at other measures of size at delivery. So a couple

1 studies looking at birth length, head circumference, or
2 ponderal index, which is -- you can think about it as a
3 measure of the leanness of the baby.
4 And these are the same three studies that also I
5 mentioned had looked at birth weight, and the -- sort of
6 the significant results sort of parallel with that. So
7 again, Lee and Philippat found -- Lee found that there was
8 a positive association between BPA level and length at
9 birth, and with ponderal index. And Philippat found that
10 there was a significant positive association with head
11 circumference, so that means higher BPA, higher on these
12 measures. And Wolff found that length and head
13 circumference were not significantly associated with BPA
14 level.
15 And then one other study I will mention is looked
16 at, actually in uterine growth, and this is Snijder. And
17 they had a subset of women in the study had three measures
18 of BPA, one in each -- one measure of BPA in each
19 trimester. And so they basically looked at the growth
20 rate across gestation in these women. And they found a
21 significantly slower rate of growth among these women
22 where they had three samples from throughout pregnancy.
23 So that is -- that is basically the sum of the
24 literature on humans that I have to summarize, and I'm
25 happy to stop there.

CHAIRPERSON GOLD: Okay. Does the Panel have any questions for Dr. Carmichael?

Okay. Well, I'll take it from here.

I took a similar but somewhat different tack in reviewing the papers that were before us with regard to the human studies. I was sort of mostly looking for consistency, so I didn't totally discount the cross-sectional studies. Although, I think the cautionary notes that Dr. Carmichael mentioned in the beginning are completely appropriate.

What I did instead was to sort of make a three-point ranking of the quality of the studies as I was going through them, and then looked sort of for consistency at -- by outcome. So I organized the papers by outcome and then looked at consistency across the studies.

But for the human studies, a little bit unlike the animal studies, there weren't -- sometimes we only had one study to look at. And so then consistency doesn't really make too much sense, because you only have the one. So I focused on when we had more than one study for a given outcome to look at. And roughly for about half of the outcomes, maybe a little less, we had more than one, but I didn't restrict myself to the cross-sectional ones. I included -- I didn't exclude the cross-sectional ones.

I included them and the longitudinal ones, but gave the longitudinal ones sort of more of a positive score in than cross-sectional ones.

So for several outcomes, we did have more than one study. So particularly if we're looking at estrogen levels, estradiol. There were several -- there were four human studies and two of these were of fairly high quality and found a significant negative association with BPA exposure. And when I said they were of fairly high quality, I thought they had adequate sample sizes and had a longitudinal design, just to give you an idea.

There were also some experimental studies on human tissues and so forth. And we had several of those that were of relatively good quality and design with a reasonable sample size. And three that I would say were of moderate quality, and also found a decrease in estrogen levels with BPA exposure.

So to me in the area of hormone production, there were fairly consistent results across a number of studies, resulting in decreased estrogen with BPA exposure.

There was one study of steroid gene expression, specifically CYP 19 expression, and found no association. But this particular study was relatively poor quality. It was a small sample size. But there were a number of experimental studies that looked at steroid gene

1 expression and did find an association either with a
2 steroid or steroid receptor expression, suggesting that
3 BPA may affect gene expression and thus potentially
4 steroidogenesis, which would be consistent with the
5 previous studies I mentioned on hormone production.
6 Let's see, Dr. Carmichael mentioned the oocytes
7 retrieved, so I don't think I really need to repeat what
8 she has said. There was one that was longitudinal, a good
9 sample size and found a negative population relationship.
10 She also mentioned birth weight, which again, I
11 don't need to repeat what she said except that I would say
12 there were -- I found six studies and they were sort of
13 all across the map in their findings. So I didn't see
14 consistency there.
15 She mentioned the study about endometriosis. I
16 don't need to repeat that, and precocious puberty, and the
17 fetal growth. There were two human studies on spontaneous
18 abortion, both of modest quality, I would say. And one
19 found a positive association and one found no association.
20 So lack of consistency I would say on that outcome.
21 Gestational duration, there were two studies,
22 both of marginal quality and both finding a significant
23 negative association. And on the experimental studies in
24 humans -- in human tissues, there were two studies of,
25 what I would say, moderate to poor quality that found a

negative association with follicular growth or formation. But due to the less desirable study quality kind of makes this relationship uncertain.

Now, for those outcomes for which there was only one study available, I put more emphasis on the quality of the study design, and the implementation and analysis was important, because one high quality study could carry a fair amount of weight than one, you know, poorly conducted study.

But for the remaining outcomes, there was only one study in humans -- for which there was only one study in humans. The majority were of modest quality, which made the possibility of making conclusions very tenuous for all except I think two of the outcomes. There was thyroid function and pre-term birth in humans. So the one relatively good study of thyroid function found a significant negative effect on thyroid function, specifically reduced maternal thyroxine levels and reduced TSH in boys.

I would say that's suggestive requires confirmation. And there was a recent study on pre-term birth, but did not find a significant association.

So I would say, in conclusion, that for me the human studies and a little bit of what we heard about the animals this morning, the animal experiments, that there

1 does seem to be an adverse effect on steroid production by
2 BPA, especially for estradiol and perhaps steroid gene
3 expression.
4 And the relative consistency of these findings in
5 humans and animals and the relatively high quality of some
6 of the studies in humans on the effects of estradiol
7 production underscore the importance of these findings.
8 So I'll stop there and see if there are any
9 questions from my Panel members?
10 COMMITTEE MEMBER CARMICHAEL: I have a question.
11 CHAIRPERSON GOLD: Yep.
12 COMMITTEE MEMBER CARMICHAEL: The studies on
13 estradiol, so what was the timing of those, do you recall?
14 CHAIRPERSON GOLD: So several of those were --
15 they were done with IVF, and so they were looking at peak
16 estradiol, so right around the time of ovulation, just
17 before.
18 COMMITTEE MEMBER CARMICHAEL: Are there any
19 that -- so I -- those are the two I'm familiar with. So
20 what was -- is there a timing to the other ones? Were
21 they non-IVF patients or...
22 CHAIRPERSON GOLD: Let me see. Obviously, I have
23 the Bloom and the Ehrlich, and the Mok-Lin was a subset of
24 the Ehrlich study, so -- also those were timed. The
25 Romani study I don't -- that was an in vitro study, so

that was not like, you know, really what I would call an epidemiologic study, but it used human cells.

Does that help?

COMMITTEE MEMBER CARMICHAEL: That helps.

CHAIRPERSON GOLD: And it's a good question, because over the menstrual cycle, estrogen varies greatly. And so it depends when you're measuring them, and if they were all measure -- most of them were focused on peak estradiol. So they're trying to get it right around mid-cycle.

COMMITTEE MEMBER CARMICHAEL: Okay.

CHAIRPERSON GOLD: Other questions, comments?

Dr. Pessah.

COMMITTEE MEMBER PESSAH: You mentioned that the changes in steroid receptor expression were stronger in the studies you reviewed. Where were those measurements made in which --

CHAIRPERSON GOLD: What do you mean?

COMMITTEE MEMBER PESSAH: -- which tissue, blood levels, or -- I mean, because obviously there had --

CHAIRPERSON GOLD: I'm sorry. I don't recall, but if you want, I'll take a break.

Sorry. Dr. Baskin.

COMMITTEE MEMBER BASKIN: Blood and urine. Blood -- or mostly urine.

CHAIRPERSON GOLD: In urine mostly. And he's also going to comment more on steroidogenesis in a moment, right?

COMMITTEE MEMBER BASKIN: (Nods head.)

CHAIRPERSON GOLD: Okay. Thank you.

So we'll come back to it.

Other questions or comments?

Are we ready for the next topic then?

So, Dr. Baskin, you're going to lead us through androgen, steroidogenesis, and exposures in females that might affect males.

COMMITTEE MEMBER BASKIN: I kind of wanted to give a global summary, because a lot of the papers have been discussed in detail. But I guess we're concerned that BPA has adverse effects on human female steroidogenesis slash it's hard to separate that in my mind from development. And this rests on the substantial literature of BPA and listing developmental defects in both female as well as male laboratory animals.

And specifically, in the animals BPA elicits uterine hyperplasia, altered uterine gene expression, clefting of the clitoris, early vaginal opening, irregular estrous cycles, persistent vaginal cornification, and, as was highlighted today, which I think is the strongest animal evidence, multiple ovarian abnormalities.

I mean, BPA was designed to be a estrogen, and it turns out it's a weak estrogen, but nevertheless it's an estrogen. And it seems to act both through estrogenic -- the estrogen receptor as well as there seems to be non-estrogenic pathways.

It's also noted that there's multiple adverse metabolic effects and behavioral abnormalities. And I guess the papers that I would cite that supports the statements I just stated would be the Rochester paper, which is a review paper, that was already alluded to, the Vandenberg paper from 2013, and the Anjum paper from -- in Reproductive Toxicology from 2013.

So despite this large body of animal research on BPA showing changes not only in steroidogenesis/female reproductive abnormalities, my reading the literature is that there is no direct evidence that BPA actually affects development in the human fetus at any, dose in fact, not just the high doses, but at the low doses, which I think is kind of an important point. And I'm not an epidemiologist, and I appreciate Dr. Carmichael's presentation.

But nevertheless, the human studies, the major impediment is that they're not control studies of exposure to BPA, since these ethically couldn't, can't, and won't be done on pregnant women or children for obvious reasons.

Thus, the inferred adverse health effects of prenatal BP exposure in humans are based solely on animal studies, which is obviously very relevant here, and correlation of epidemiologic studies in the human population.

So a major concern is there's substantial evidence of widespread human exposure to BPA. In other words, we've all got it in our bodies. There's no question about that in my mind. Whether it's dangerous or not is what's really under consideration here. So BPA has been detected in air, dust, urine, breast milk, pregnant women, amniotic fluid, umbilical cord blood, placental tissue, human fetal tissue, including the liver. And so there's no question that we're exposed to this.

Again, I would emphasize in the human studies there's really no control group. In other words, there's no population I know of, at least here, that has not been exposed to BPA. So it could be good for us. We don't really know.

So why -- so I would summarize that while there are certainly plausible links to BPA being adverse in humans, the epidemiologic studies are suggestive, and most of the factual material is in the animal studies.

So then I would focus that animal studies are relevant, and that the key points that I found in the

1 literature is that at doses lower than what is recommended
2 BPA exposure, which is less than 50 micrograms per
3 kilogram per body weight, there are a number of properly
4 done scientific studies that were alluded to already by my
5 colleagues on the panel that clearly showed abnormalities
6 in steroidogenesis, specifically the ovary, okay, and
7 female reproductive tract.
8 And this implies to me that the present
9 documented level of safe exposure of BPA should be --
10 simply be revisited. And I'm going to leave it at that.
11 CHAIRPERSON GOLD: Any questions for Dr. Baskin?
12 Okay. Did we want to take a short break to
13 organize the public comments. So this is your last chance
14 I think to get the blue cards in if you would like to
15 speak. And then we're going to organize them. We're just
16 taking a really short break, like two minutes, and then
17 we'll come back.
18 (Off record: 1:14 PM)
19 (Thereupon a recess was taken.)
20 (On record: 1:16 PM)
21 CHAIRPERSON GOLD: Okay. I think we're ready
22 to -- so first, Ms. Monahan Cummings is going to talk a
23 bit about the timing and then we'll go from there.
24 CHIEF COUNSEL MONAHAN CUMMINGS: Good afternoon.
25 I just wanted to let you know that we do have quite a

number of folks that are planning to speak today. As Dr. Gold mentioned, there were three groups that asked for time prior to the meeting. And Dr. Gold went through those requests. Excuse me. Our first presenter is going to be from the NRDC. They requested 15 minutes and that was granted. The other two groups asked for considerably longer periods of time, and Dr. Gold determined that 20 minutes each for the two groups of ACC and ACMI would be appropriate. The rest of the commenters are all individuals. And so our plan today is to give each individual five minutes per person, other than the group presentations.

This room is equipped with a timer that is on the podium. And so our staff will be setting the timer for you as speakers. And we appreciate it if you would keep an eye on the timer. It will beep when you're done. I'm not sure whether or not it will do that just prior to the end of your time, but you can keep track of it by looking at it.

We do need you to stay on time, because we do have lots of speakers today. And to the extent that you agree with prior speakers, you're more than welcome to say I agree with a prior speaker and not repeat what they had to say. That can be real helpful just in terms of timing.

CHAIRPERSON GOLD: Thank you very much.

So first, we'll hear from the NRDC. They have a coordinated group presentation for 15 minutes.

Could you also please introduce yourselves as you come up?

Can I just clarify one thing with you. We have three cards for the NRDC, but there are only two of you standing up there, so is the third person joining you or --

MR. KAR: No, I think it's going to be the two of us.

CHAIRPERSON GOLD: Just the two of you. Okay. Thank you.

(Thereupon an overhead presentation was presented as follows.)

MR. KAR: Well, we could maybe go ahead and get ourselves introduced in the meantime. Thank you again for the opportunity to comment. My name is Avinash Kar and I'm an attorney with the Health and Environment Program at the Natural Resources Defense Counsel.

NRDC is a national environmental organization that advocates for policies that protect public health from harmful chemicals in the environment. NRDC has 2.4 million members and on-line activists, 380,000 of who are Californians. Funding for my work comes predominantly from private foundations and individuals who care about

environmental health. And NRDC paid for my travel here today.

NRDC strongly supports listing of BPA as a reproductive toxicant. And we'll go through our presentation in a moment. I'll let Dr. Rochester introduce herself.

DR. ROCHESTER: My name is Johanna Rochester. I'm a research associate at The Endocrine Disruption Exchange in Colorado. We're a group that works to clarify the science behind endocrine disruptors for policymakers, scientists, and the public. I've published reviews on BPA and BPA analogs, exploring the physiological actions and human health effects of these compounds. I'm here on behalf of TEDX and the NRDC. And the NRDC paid for my travel here.

Just to further introduce myself. Last year, I published a review that examined all the studies that explored BPA and health effects in humans. There were over 90 studies at the time. And 75 of them showed significant correlations. This review identified multiple adverse health effects in humans, and has been highly cited since its publication.

MR. KAR: What we plan to cover -- this is the outline for it. We just want to cover what -- what is the criteria for listing, what exactly is getting listed and

the scientific evidence as they match up to that criteria. Our intent is not to revisit the science at the level of detail that has been discussed already today. It's to show how well the scientific evidence maps to the criteria which guide the Committee's evaluation of the chemical.

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MR. KAR: So what is listed? You know, as you know, Proposition 65 lists both reproductive toxicants and carcinogens. And specifically, we're talking about female reproductive toxicity. The two impacts that are contemplated by the criteria are adverse effects on reproductive structure or function and impaired reproductive performance. And those are the two impacts that we will focus on -- those two sets of impacts that we'll focus on as we go through this.

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MR. KAR: The first set of impacts, of course, will be female reproductive toxicity -- I'm sorry, adverse effects on reproductive structure or function. But we want to point out before that, that what is required for a listing is that one of these two criteria has to be met. It's either sufficient evidence of reproductive toxicity in humans or sufficient evidence of reproductive toxicity in animals, either one of these is sufficient for a listing.

And even one study can -- even one strong study can be sufficient evidence. Although, of course, multiple studies increase the confidence.

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MR. KAR: Other considerations, as you discussed earlier today were biological plausibility and statistical considerations, and again, focusing on adverse effects on reproductive structure or function first.

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MR. KAR: There are, of course, multiple different reproductive effects in women. For -- to simplify the presentation today, we'll focus on one of these reproductive effects, the disrupted ovulation oocyte maturation, as an example, to illustrate the strength of the literature and that the criteria for listing have been met.

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MR. KAR: The criteria explicitly define adverse effects on reproductive structure and function to include several different facets. One, genetic damage to the ovum or its precursors, alterations in ovulation or the menstrual cycle, and/or menstrual disorders, and impaired or altered endocrine function, among others. Evidence of any one of these effects is sufficient for listing.

Dr. Rochester will focus the -- will discuss the

scientific literature, demonstrating these effects of BPA, focusing on some key studies demonstrating these effects.

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DR. ROCHESTER: So we'll start with the human studies. These studies highlighted included several populations of women that were treated at fertility clinics, as we've already discussed. BPA was measured in the blood and urine, and exposure to BPA was correlated with these outcomes when the subjects underwent fertility treatments.

For the disruption to the ovum, BPA was associated with a reduction in mature oocytes in women, as well as reduced probability of oocyte fertilization. BPA was also linked to alterations in ovulation. When ovulation was induced by reproductive hormones, higher BPA levels were associated with poor ovulation response.

BPA was also associated with less estrogen during the stimulated ovulation, an example of disruptive endocrine function. These studies are particularly strong, because they're repeated by several independent research groups, and they were prospective cohort studies, which are able to correlate the time of disruption to exposure.

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DR. ROCHESTER: I'm going to give a little

background about normal reproductive endpoints in humans in relation to these studies. For normal oocyte development, oocytes go through stages of splitting the chromosomes and dividing. This is called meiosis, and there are two phases of meiosis.

All the oocytes a woman has have developed by puberty, but they are paused at a certain stage of development in Meiosis II until fertilization. If the oocytes have not reached a certain stage by this time, they will not be viable for fertilization. The previously mentioned studies found that there were significantly more oocytes that had not reached that normal stage in the woman exposed to higher BPA.

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DR. ROCHESTER: For normal ovulation in humans, reproductive hormone signals from the pituitary gland, which is signaled by the brain, act on the oocytes and the ovaries. The ovaries in turn cause the oocytes to release estrogen which acts back on the brain. Ovulation can be induced by exposing women to a reproductive hormone, and this is routinely done during fertility treatments.

This stimulation causes multiple oocytes to be released from the ovaries, as well as a surge of estrogen produced from the oocytes. In the previously mentioned studies, women with higher levels of BPA had poorer

ovarian response, which means they had a reduced number of eggs released and less estrogen produced by the stimulation.

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DR. ROCHESTER: The disruption of the oocytes and the other toxic effects on reproductive structure are supported by animal research. Mice and monkeys both showed disrupted oocyte development with BPA exposure. Particularly, they showed disruptive meiosis in oocytes, similar to effects in humans. BPA exposure in mice cause a delayed disrupted estrous -- delayed and disrupted estrous cycle, which is equivalent to ovulation in humans; BPA impaired endocrine function in mice by affecting the number of estrogen receptors in the brain.

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DR. ROCHESTER: There's also a lot of mechanistic evidence in cells and animals that support the biological plausibility of BPA being toxic to reproductive structure in humans and animals. The disruptions in meiosis in human and animal oocytes have been explored in several in vitro studies. In the ovum, BPA causes changes in the spindle fibers, which are crucial for meiosis.

There are also mechanistic studies that support the other criteria. It was shown that the disruption of estrous by BPA in mice is mainly due to disrupted ovaries.

Lastly, it's well known that BPA can interfere with endocrine function by binding to estrogen and androgen receptors.

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MR. KAR: Now, we will turn to the second set of impacts, which constitute female reproductive toxicity that has impaired reproductive performance.

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MR. KAR: Again, the criteria defined impaired reproductive performance to include increased pregnancy wastage, inability or decreased ability to conceive, and adverse effects on sexual behavior, gestation, lactation, fertility, onset of puberty, parturition or premature reproductive senescence. Any one of these effects is sufficient for listing.

Dr. Rochester will again discuss the scientific literature documenting these effects of BPA focusing on some of the key studies.

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DR. ROCHESTER: Again, I'll begin with all of the studies in humans. Higher BPA exposure has been linked to increased rates of miscarriage in two different populations of women. And BPA exposure has been associated with increased implantation failure. Also, women with higher levels of BPA had a higher probability

of being infertile. Lastly, higher levels of BPA in women were also associated with increased rates of premature delivery.

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DR. ROCHESTER: The animal data also supports a disruption of reproductive performance. In mice, BPA caused pregnancy failure and implantation failure. And in rats, BPA caused fetal death and fetal malformations. BPA exposure caused accelerated infertility in female mice with aging of the females.

BPA has been shown to cause changes in sexual behavior in female mice. And also in mice, lactating dams exposed to BPA had a reduced rate of growth of their pups, which was due to less milk being produced from the dams.

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DR. ROCHESTER: Many mechanistic studies support these findings of disrupted reproductive performance. BPA has been shown to be toxic to embryos in vitro. In animals, BPA disrupts the development of the reproductive tract, which can lead to the inability to conceive. BPA has also been shown to alter the release of prolactin in vitro which is a hormone involved in lactation, and thus disrupt milk production.

It was shown to permanently disrupt the normal brain mechanisms that drive female sexual behavior, thus

providing a mechanism for the altered sexual behavior seen with BPA exposure.

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MR. KAR: Once again, just to come back to what is listed. The evidence -- there's sufficient evidence of one of these impacts, either adverse effects on reproductive structure or function or impaired reproductive performance, either of these in humans or animals or in combination is sufficient for a listing.

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MR. KAR: Unlike some other bodies that have reviewed BPA, as Ms. Monahan Cummings mentioned earlier today, the DART's inquiry is focused on whether there is sufficient evidence of reproductive toxicity guided by the criteria we just discussed. We believe the scientific literature demonstrates sufficient evidence of female reproductive toxicity.

Today's decision that you're going to be making reflects your independent judgment as the State's experts on the science responding to Proposition 65's specific criteria. The risk and exposure issues that may come up are addressed at a later stage in the process. The Committee will have an opportunity to review and comment on OEHHA's assessment of risk and exposure and any proposed action at that stage.

We thank you once again for your time.

CHAIRPERSON GOLD: Thank you. Could you stay for one second, please.

Are there any questions from the panel of the NRDC?

Okay. Thank you very much.

Okay. Next, we'll hear from the ACC. You have 20 minutes as a coordinated group of presentations.

(Thereupon an overhead presentation was presented as follows.)

MR. LANDFAIR: Just to clarify, Dr. Gold. We'll be followed by ACMI, which is also given 20 minutes, and we've coordinated our two presentations.

CHAIRPERSON GOLD: That's correct. Thank you.

MR. LANDFAIR: Thank you. While she's setting the timer, if I may, I just can't begin without acknowledging the announcement that was made this morning concerning Dr. Alexeeff. We know each other only professionally and usually on the opposite sides of professional disagreement. But Dr. Alexeeff has always been a true gentleman, a person who's open to discussion, to debate, who encouraged it, treated everyone with respect. When you come into a meeting like this and find he's not here, you're impressed with just how fragile and short life is. And I don't know of George's condition,

but we wish him the best, and we should all treat each other well.

Thank you. My name is Stan Landfair. Thank you. I am an attorney with the firm McKenna, Long & Aldridge. I represent the American Chemistry Council. I do not pretend to be a scientist. My role is to help our clients to articulate their issues and put this presentation and their comments together. I'll also be introducing our speakers.

So moving on to -- the best place to start is I want to thank you. I want to thank the Committee for their hard work it's obviously put in. This is one of the more exhaustive Committee reviews we've ever seen of the data we put in from of them, and we look forward to the opportunity for this discussion. We want to thank you for the opportunity for a coordinated presentation and then ACMI for working together with us.

So moving on to the introductions. We provided you with a copy of our comments bound, and I want you to be able to associate the submitter with the submission, and introduce the speakers from that. Dr. Hentges who works full time at the American Chemistry Council has worked for 15 years exclusively with bisphenol A. He's very familiar with the database. And we encourage you to ask him, as we do with all our speakers, any questions you

have regarding the data, as he's made a full-time job out of this for 15 years.

Dr. Goodman, in addition to being a epidemiologist -- in addition to working for the consulting company Gradient, also is an adjunct professor at Harvard University. Our next speaker, Anthony Scialli, in addition to be a private consultant and a medical doctor is also an adjunct professor at Georgetown University Medical School and a full-time professor at George Washington Medical School.

And Jay Murray probably needs the least introduction, but we want to point out that he was one of the first -- he was a member of the first DART IC. And our colleagues from ACMI will introduce themselves.

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MR. LANDFAIR: With that said, I want to move on ever so briefly to the issue of the standard for listing. Carol, of course, was correct, perhaps a mind reader or a predictor of the future in the fact that we have to discuss this. We can't avoid discussing this, even if some people would prefer we not.

We've heard the recitation of the standard many times. We want you to know that's in the statute. It wasn't a lawyer like me who made this up. This is the reason for it. It's what the statute calls for. The

implementing regulations call for it. And what they call for is a determination of whether or not a chemical has been clearly shown.

Your criteria are your criteria, but that's where the idea of the weight of the evidence comes from. And we ask you, when you evaluate a data for -- chemical for any particular endpoint, including one these -- some of these subendpoints, ask yourself whether we've acknowledged and reviewed all of the evidence and can conclude, in our own intellectual honesty, that the weight of the evidence supports a conclusion on any particular endpoint.

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MR. LANDFAIR: Now this comes up so often, what does it mean to be clearly shown?

The debate between whether it's a scientific standard or a legal standard, I think that's -- it's an issue I don't need to discuss. These are common words. They know things we all know what they mean, show clearly. If we need to treat them as a legal phrase, show clearly equals prove in the legal thesaurus, and in a non-legal phrase. This is the English language, the one we all speak. Show clearly equals prove, and there are many known synonyms for it.

--o0o--

MR. LANDFAIR: So the reason -- one of the

reasons we have to discuss this is because frequently we get comments from advocates for listing, or sometimes scientists, or sometimes frankly members of the Panel who would say, well, the data suggest, or it's likely to be a reproductive toxicant, or it's likely to be a cause of this. I have concerns. I want to err on the side of health and safety, the precautionary principle. None of those articulate the standard.

And the reason the standard is so rigorous is because Proposition 65 -- I'm not going to talk about the consequences, but Proposition 65 is sort of a blunt instrument as a regulatory tool, and we need to make sure we adhere to the standard for identifying a chemical on the Prop. 65 list.

So with that, I'm going to leave this up as sort of the agenda and score card as these other people speak.

Steve.

DR. HENTGES: Good afternoon, and I'd like to start simply by seconding what Stan said about Dr. Alexeeff. We all -- not hearing me.

CHAIRPERSON GOLD: Try getting close.

DR. HENTGES: Okay. I'll lean in.

We all -- thank you. We all have him in our thoughts and prayers now and hope the best for him.

So back to the topic of the day, which seems a

1 little small in comparison. There are three things that I
2 want to talk about. I'm Dr. Steve Hentges with the
3 American Chemistry Council.
4 Three things that I want to touch on. First is
5 FDA's assessment of BPA. You know that FDA released very
6 recently in November of last year --
7 CHAIRPERSON GOLD: Wait a second. I think we're
8 having a little trouble hearing you. Is your green light
9 on there on your microphone.
10 Sorry?
11 It's on. Just checking. Maybe if you can move
12 it closer to you, that would help. Thank you.
13 DR. HENTGES: Okay. So you know that FDA
14 recently released their comprehensive safety assessment of
15 BPA. Their overall conclusion on safety, you've seen this
16 in the short letter that you received from FDA's chief
17 scientist, they conclude that BPA is safe at the current
18 levels occurring in food.
19 But don't be deceived by the brevity of the
20 letter. There's a lot behind it. FDA has conducted a
21 very thorough and well-documented hazard identification
22 process. I think you've seen the documentation on that
23 many hundreds of pages. FDA applied well-defined hazard
24 identification criteria to evaluate individual studies.
25 Their hazard identification criteria and process was

separate and distinct from the risk or safety assessment. They had separate criteria and a separate process for the risk or safety assessment.

Everything is thoroughly documented in several lengthy memoranda, which FDA considers as the current state of the science evaluation and hazard characterization of BPA.

The assessment was conducted by a broad cross-section of scientific experts from throughout FDA. In the last memorandum, there were 38 scientific experts that were co-authors. And hazards -- after evaluating individual studies, hazards were identified by the weight of the evidence, which is the same way that the DART Committee evaluates hazards.

The bottom line from FDA, as far as hazards, or in particular regarding reproductive toxicity is they did not identify reproductive toxicity, either male or female, as a hazard of BPA. Now, that's partly significant because FDA is designated, for purposes of Proposition 65, as an authoritative body. And what that means in practice is that had FDA identified BPA as a reproductive toxicant, OEHHA could have proposed listing BPA simply based on the FDA assessment, and we wouldn't be here talking about it today at all, but they didn't. They did not find reproductive toxicity as a hazard of BPA.

1 That leads to my second topic that I want to
2 touch on which is FDA's research on BPA. Beginning in
3 2009, FDA, in conjunction with the National Toxicology
4 Program, designed a comprehensive research program on BPA
5 to answer key scientific questions and resolve
6 uncertainties about the safety of BPA.
7 And, in particular, they aimed to resolve
8 uncertainties that were identified in the 2008 NTP report.
9 That's the one that Jim Donald mentioned was a key
10 document back in 2009. The studies are funded by NTP and
11 conducted at the National Center for Toxicological
12 Research, NCTR, in Arkansas.
13 To date, 17 studies published in the
14 peer-reviewed scientific literature included are both
15 toxicity studies, as well as a comprehensive set of
16 pharmacokinetic studies, both in rodents and in non-human
17 primates. Dr. Scialli will discuss the key toxicity study
18 from that program when he steps up to the microphone in a
19 few minutes.
20 I'll just mention that that study is probably the
21 largest toxicity study ever conducted on BPA. It was also
22 briefly mentioned in the letter from FDA's chief
23 scientist, where they stated that the data do not support
24 BPA as a reproductive toxicant.
25 So that is now my segue to the pharmacokinetic

1 studies thatI want to touch on as my third topic. What do
2 they tell us in particular about biological plausibility?
3 As you know from your Committee guidance
4 criteria, metabolic and pharmacokinetic data can increase
5 or decrease the confidence for classification of an agent
6 as a reproductive toxicant. And as with just about
7 everything with BPA, there's an abundance of
8 pharmacokinetic data available. And of particular
9 importance are the set of well-designed and coherent
10 studies conducted at NCTR.
11 Overall, the pharmacokinetic studies suggest low
12 biological plausibility for BPA as a reproductive toxicant
13 in humans. And with limited time, I'm just going to give
14 you some headline conclusions that come out of these
15 studies. In general, humans efficiently metabolize and
16 rapidly eliminate BPA after oral exposure, which is the
17 most relevant for humans through the diet.
18 What happens after oral exposure is BPA undergoes
19 efficient first-pass metabolism, both in the intestine and
20 then in the liver before anything enters systemic
21 circulation. Because of the efficient metabolism, the
22 systemic bioavailability of BPA is quite low, less than
23 one percent of the administered dose goes into systemic
24 circulation. And the half-life of BPA is quite short,
25 terminal half-life about five to six hours, meaning that

1 BPA is eliminated, within the day of exposure. It's
2 eliminated in urine.
3 Pharmacokinetic profile of BPA is similar for
4 pregnant and non-pregnant females, in monkeys that is.
5 And in both cases, internal exposure is quite low, and in
6 particular internal -- very importantly, internal exposure
7 to the fetus is actually less than the mother.
8 There are several studies now in human
9 volunteers, pharmacokinetic studies, with controlled
10 doses. The results of those studies are remarkably
11 similar to the pharmacokinetic studies in monkeys.
12 Regarding biological plausibility, another
13 important point is that the metabolites of BPA, which
14 predominantly is what goes into circulation are not
15 estrogenic. It was pointed out earlier that BPA well
16 known to be weakly estrogenic, metabolites are not, which
17 suggests that BPA is not likely to cause estrogenic
18 effects after oral exposure.
19 Now, there's three last points that I want to
20 distill out of the pharmacokinetic data that really touch
21 on things that you discussed this morning. First, is that
22 non-oral pharmacokinetics are significantly different from
23 oral. And this is important because quite a few toxicity
24 studies are conducted with non-oral routes of exposure,
25 subcutaneous being the most common of those.

1 For example, I think the sheep studies that were
2 mentioned this morning were probably all subcutaneous
3 exposure. So what happens is that with non-oral exposure,
4 the efficient first-pass metabolism is bypassed, resulting
5 in significantly higher bioavailability of BPA circulating
6 parent BPA.
7 And as result of that, toxicity studies with
8 non-oral exposure will be of limited relevance for human
9 hazard assessment. The second point to distill out is
10 that human and non-human primate neonates have metabolized
11 BPA very efficiently. Only minimal pharmacokinetic
12 differences between adult and neonatal monkeys, in both
13 cases very low bioavailability, after oral administration,
14 there are no age-related changes in internal exposures.
15 That's been corroborated in two observational studies on
16 human neonates, as young as three days of age.
17 And the significance of this is that there are
18 significant age-related changes in developing rats.
19 Neonatal rats, or more generally rodents, are well known
20 to have a deficient ability to metabolize BPA. And what
21 that tells us, this is really FDA's conclusion, is that
22 toxicity studies in rodents from early postnatal exposure
23 are likely to overpredict the effects on primates of the
24 same age.
25 And then the last point that I want to make has

1 to do with something that Dr. Pessah, and I think Dr.
2 Plopper may have touched on very briefly, regarding
3 circulating levels of BPA in the human population. And
4 there are reports, I think as, in particular, Dr. Pessah,
5 that you mentioned that report nanomolar levels of parent
6 BPA, free BPA in human blood.
7 But there's now growing awareness that that data
8 is likely to be a result of contaminations. And I'll
9 mention three things very quickly before I use up
10 everybody's time here. One is a paper from CDC
11 researchers published in 2013 on potential external
12 contamination with bisphenol A during biomonitoring
13 analysis. A second is a letter to the editor from Calafat
14 et al. Antonia Calafat is a well known researcher and
15 biomonitoring expert at CDC. The title tells it all,
16 "Misuse of Blood Serum to Assess Exposure to Bisphenol A
17 and Phthalates". And they state for the reasons discussed
18 in the paper, urine is the best matrix for epidemiological
19 assessment of exposure to BPA.
20 And there's a few others I could go on and give
21 examples from FDA's research, in particular the
22 pharmacokinetic data, that further supports that the
23 levels -- the nanomolar levels of BPA in human blood are
24 really implausible. So that's -- I think I need to stop
25 here and maybe give you a chance for a quick question, if

you have one?

CHAIRPERSON GOLD: I think you should keep going and maybe we'll come back. Can you hold it.

DR. GOODMAN: Thank you. I want to talk about epidemiology briefly. In 2009, the DART Committee determined that study design limitations led to limitations and study findings -- Oh, sorry. I'm Julie Goodman. I'm third on the list from Gradient -- that there have been many, many new studies conducted since 2009, but all of them have the same limitations, the same uncertainties as those conducted before.

And Dr. Carmichael mentioned several of these limitations, but even talking about these limitations, she focused on the higher quality studies. And granted, among all the studies, some of them are certainly higher quality than others. But as a whole, they all have these limitations, and even the higher quality ones are not sufficient to base conclusions on.

You know, just for example, it is true two BPA measurements are probably better than one, but that's still not good enough. Exposure levels are so small, often straddling the limit of detection in studies. And the ranges are so small, that the probability of exposure misclassification or exposure measurement error are so high, you really don't know how to interpret those

1 results, even in those studies with two measurements or
2 three.
3 The next point is even if you -- you know,
4 setting this aside, there's been a lot of discussion of
5 studies of hormone expression -- or hormone levels and
6 gene expression. And certainly, you know, changes in gene
7 expression or hormone levels could potentially lead to
8 reproductive effects, but in and of themselves, those are
9 not reproductive effects. They are not adverse effects.
10 And without information on whether the particular -- the
11 degree of increase in hormone levels or decrease or the
12 degree of the increase in gene expressions or particular
13 genes, if that hasn't been shown to be associated with
14 reproductive effects, then you cannot conclude that those
15 are evidence for reproductive effects.
16 Finally, you know, I mentioned the DART Committee
17 in 2009, we also have NTP CERHR in 2008, FDA in 2014, and
18 the European Food and Safety Authority in this year, all
19 reviewed these epidemiology studies in detail, and all
20 concluded that there were too many limitations and too
21 many uncertainties to draw conclusions. And so because of
22 this, you cannot -- these studies are not adequate to
23 determine whether or not bisphenol clearly shows -- or the
24 evidence clearly shows causation, either with themselves
25 or as support for animal studies.

Thank you.

DR. SCIALLI: Hello. My name is Tony Scialli, and I'm an obstetrician/gynecologist and reproductive toxicologist. In fact, I was the founding editor, and for 17 years, the editor-in-chief of the Journal of Reproductive Toxicology, in which you found some of the papers that you reviewed for today.

I talk to patients and -- I talk to patients who are concerned about exposures and patients who are concerned about fertility often coming to ask me why they haven't gotten pregnant?

What I'd like to review for you briefly are the conventional experimental animal studies, which I so far haven't heard mentioned except by my colleagues who just spoke. There are seven conventional studies. And I like considering the conventional studies, by which I mean studies that are often used for regulation, because they have controlled exposures. They evaluate relevant endpoints, largely apical endpoints. And they can be carefully constructed and evaluated to answer some of the questions that are raised by the mechanistic studies that you've reviewed.

I have to wonder if, in fact, bisphenol A causes these abnormalities in meiosis and in reproductive success, why haven't any of the seven studies that have

used conventional design show it?

Now, there are studies that were done by the time of the 2009 review. I'd like to focus on one study that was done since that time. That's the study that was done at NCTR with the support of the National Toxicology Program. The toxicology paper from that study was published by Barry Delclos et al. in 2014. There is also, however, a study from -- excuse me, a paper from that study by Camacho et al. that looked at gene expression endpoints, and was negative. There was a study by Churchwell that looked at the dosimetry. This study --

CHAIRPERSON GOLD: I want to remind you, you have less than 30 seconds.

DR. SCIALLI: We're going -- I'm sorry, we've arranged to combine our time.

CHAIRPERSON GOLD: Have you switched to the ACMI now?

DR. SCIALLI: Excuse me?

MR. LANDFAIR: ACC will finish and then we'll hear from ACMI.

CHAIRPERSON GOLD: Okay.

MR. LANDFAIR: Thank you.

DR. SCIALLI: Thank you. So the Delclos study involved dosing of Sprague-Dawley rats from -- thank you -- by gavage from gestation day 6 to postnatal day 90.

1 There were dose levels that ranged from 2.5 micrograms per
2 kilogram body weight per day to 300,000 micrograms per
3 kilogram body weight per day. There were two positive
4 controls with two different doses of ethinyl estradiol and
5 two negative controls, one naive control and one
6 vehicle-treated control.
7 Except for effects that occurred at manifestly
8 systemically toxic dose levels of 100,000 and 300,000
9 micrograms per kilogram per day, there were no adverse
10 reproductive effects. There were no effects on
11 histopathology at 90 days of age of the ovary, including
12 follicle counts, corpus luteum counts, uterus, mammary
13 gland. There were no abnormalities of hormone levels.
14 So I would suggest that this is an important
15 study to consider when considering the entire body of
16 literature as to possible reproductive effects of
17 bisphenol A.
18 Thank you very much.
19 CHAIRPERSON GOLD: Can I just say we've had a
20 request for a five minute break. So we'll -- you have 18
21 and a half minutes when we come back, is that okay?
22 MR. LANDFAIR: 18:47 when you said excuse me.
23 (Laughter.)
24 CHAIRPERSON GOLD: I won't argue with you if
25 you'll give us a five-minute break.

(Laughter.)

(Off record: 1:56 PM)

(Thereupon a recess was taken.)

(On record: 2:04 PM)

CHAIRPERSON GOLD: Okay. Before you start, we need a point of clarification up here. So we gave 20 minutes to the ACC and 20 minutes to the ACMI. Have you combined those to 40 minutes? Is that's what happened? I'm just checking.

MR. LANDFAIR: In effect, yes.

CHAIRPERSON GOLD: Okay.

MR. LANDFAIR: My understanding --

CHAIRPERSON GOLD: Okay. So you're on your second 20 minutes, and I'll add 10 seconds or 12 seconds to what's on the clock, okay?

MR. LANDFAIR: That would be great, and thank you for your understanding and hope we did not misunderstand.

CHAIRPERSON GOLD: Okay.

DR. MURRAY: Thank you, Dr. Gold. I'm Dr. Jay Murray. And first, thank you for your diligence in reviewing all these studies. I'm going to briefly summarize our comments on the unconventional studies. And I call them unconventional, because that's the term that NTP used to describe these studies that have unconventional experimental designs or protocols that have

1 not been validated.
2 Most of these studies, as you know, use very low
3 doses, doses that are typically orders of magnitude below
4 the NOELs in the conventional toxicity studies. And the
5 unconventional -- what I'm referring to as the
6 unconventional studies certainly have value for generating
7 hypotheses, but it's important to test those hypotheses in
8 studies that have adequate designs and factors.
9 And, you know, things like adequate numbers of
10 animals, more than a single dose level, and a route of
11 exposure that is relevant. You heard from Dr. Hentges how
12 important it is to distinguish between studies where the
13 compound was given parenterally either subcue, I.P., in an
14 implant versus oral.
15 And, you know, some of you know early in my
16 career, I worked for a pharmaceutical company that was one
17 of the companies that pioneered the development of
18 synthetic estrogens in the birth control pill. And one of
19 the challenges was to get past the metabolism in the GI
20 tract and the first-pass effect in the liver, because
21 there were a number of estrogens that didn't work when you
22 gave them orally. It was a challenge developing estrogens
23 that could be given orally that had that therapeutic
24 efficacy.
25 So, in general, the results of these studies have

not been replicated. And in the limited cases where attempts have been made to replicate the results, they often end with conflicting results, conflicting among the unconventional studies, and certainly conflicting with the results of the guideline or conventional studies.

So as you know, it's important to weigh the consistency, the evidence, as well as the strengths and limitations of the individual studies. Many of the unconventional studies have looked at things like estrogenic activity gene expression studies. And it's important to look at those things, but it's also important to keep in mind that that's mechanistic information that may be relevant for any demonstrated adverse effect on female reproduction.

But, in my opinion, the mechanistic studies alone are not enough. You have to have the demonstrated adverse effect on female reproduction. So it's instructive that no regulatory agency has relied on a NOEL from any of these studies in establishing a safe dose. These studies are consistently regarded as inadequate by government bodies FDA, NTP, CERHR, for a variety of reasons.

And in most cases, a lot of the studies that you were describing today, if you look at the FDA evaluations of those studies, many of those were determined by FDA to be of no utility for hazard identification, and they gave

their -- they gave their reasons with the limitations that the study was, that drew -- that allowed them to draw that conclusion.

And EFSA, European Food Safety Authority, made similar evaluations of many of those studies where EFSA said, you know, an interesting hypothesis, but the hypothesis needs to be tested in studies of better design or adequate design.

So, in my opinion, most of these studies would not qualify as scientifically valid testing according to generally accepted principles for purposes of Proposition 65. And even if they did, they do not provide sufficient evidence to list, in part because of the inconsistency in the results. And a number of you alluded to those, where you get, you know, a result one direction in one study and a result the other direction in another study.

So, in short, I don't believe those studies provide a reliable or adequate basis to conclude that BPA is clearly shown to cause female reproductive toxicity.

It's also important to -- you know, one of the studies that Dr. Scialli covered was the Delclos study. And the Delclos study is about as sophisticated a study as you will get. This is the one that was done by NCTR, had nine dose levels of BPA, seven of them in the low dose range, equally spaced, two negative controls, two positive

1 controls.
2 And the conclusion of Delclos -- and I'll read
3 it, because it's -- I want to make sure I quote it
4 accurately, is, "Our interpretation of the results of the
5 present study is that BPA, in the low dose region, from
6 2.5 to 2,700 micrograms per kilogram per day, did not
7 produce effects in the evaluated endpoints that differ
8 from the normal background biological variation".
9 FDA also reviewed that study separately, and had
10 their scientists peer review this study. And their --
11 FDA's conclusion was quote, "No clear treatment related
12 effects were observed in the low dose range of the study",
13 period.
14 So you've got to ask yourselves why is it that
15 we're seeing these effects in studies, but not able to
16 replicate them in the larger more conventional study.
17 So considering all the scientific evidence,
18 neither the human nor animal studies demonstrate that BPA
19 is clearly shown to cause female reproductive toxicity.
20 The most reliable animal studies show BPA is not a
21 selective female reproductive toxicant. I'm talking about
22 the conventional studies that Dr. Scialli described, and
23 the unconventional low-dose studies are suggestive,
24 certainly useful for formulating hypotheses, but you've
25 got to pursue those leads, and you've got to confirm those

hypotheses in studies of adequate design.

So, in conclusion, BPA has not been identified as a female reproductive toxicant by NTP, FDA, EFSA, or any similar authority. And finally and importantly, even if the animal studies were sufficient, which they are not, the pharmacokinetic data show that a human hazard is not biologically plausible.

I agree that you can list a chemical based on animal evidence. You don't need to establish that the compound causes female reproductive toxicity in human studies, but you have to consider biological plausibility and pharmacokinetics. It had -- the animal studies have -- you know, should indicate that it is biologically plausible. And because of the pharmacokinetics, I don't think it is biologically plausible.

So, in conclusion, the weight of the scientific evidence on BPA does not come close to meeting the clearly-shown-to-cause standard for female reproductive toxicity. Thank you.

MS. GRIMALDI: Thank you, Dr. Gold, Committee members. My name is Ann Grimaldi of Grimaldi Law Offices. I'm legal counsel for the Art and Creative Materials Institute, or ACMI. I'm here with Dr. Beth Mileson a D.A.B.T. toxicologist from Technology Sciences Group. And we appreciate this opportunity to talk with you about this

very important listing decision.

ACMI is a trade organization of approximately 190 art material manufacturers and retailers. ACMI's mission is to promote the safe use of our materials. And to that end, it sponsors a certification program pursuant to which products are evaluated by board certified toxicologists to assess acute and chronic toxicity under two federal laws, the Federal Hazardous Substances Act, and the Federal Labeling of Hazardous Art Materials Act.

If you've ever purchased crayons or a water color set or a highlighter like this, and have seen a circular symbol with the letters AP inside, you've purchased an ACMI member product that has been evaluated by a toxicologist and determined to be safe to use.

You may wonder why our material manufacturers are concerned about BPA listing here today? BPA is used in polycarbonate components of certain art materials and their packaging. ACMI's program -- certification program is based on available scientific evidence, using criteria derived from scientifically valid testing. And when there's a listing decision that does not comport with applicable listing criteria, which themselves are tied to scientifically valid testing, according to generally accepted principles, then ACMI's program -- certification program becomes compromised.

And finally, the reason for why we're here today is that ACMI members, as producers of consumer products, are in the front lines. They are the targets of enforcement actions, the soldiers in the trenches, so to speak. That's why ACMI has a strong interest ensuring that the listing decision of this chemical, or indeed any chemical, comports with the applicable listing criteria.

And that's why Dr. Mileson and I are here today, to convey this important message that listing decisions do have consequences. It is the -- a listing decision is the first step in a sequence of events that leads to the transmission of warnings, and to enforcement actions.

And I know that you are not concerned here today about enforcement actions, who gets sued for what under Proposition 65, but you are concerned with ensuring that the standard for listing is met. And you should be concerned with the public health implications of companies transmitting warnings for chemicals whose listings do not comport with the listing criteria.

And the integrity of Proposition 65, the entire law, the way it's implemented and enforced, in this first critical threshold step, depend on strict adherence to the clearly-shown-to-cause standard and the related regulatory listing criteria. The standard and the criteria not met with BPA, and BPA should not be recommended for listing.

I now yield the floor to Dr. Mileson.

DR. MILESON: Thank you, Ann. As Ann said, I'm Beth Mileson. I work for Technology Sciences Group, and I'm here to talk about BPA on behalf of ACMI.

A little shorter than that.

In the listing announcement for BPA, OEHHA provided a link -- an electronic link to a recent article -- a summary review article on BPA and reproductive health that updated experimental and human evidence over the years from 2007 to 2013.

The review article by Jackie Peretz and her colleagues summarized recent literature on BPA, and concluded that there was strong evidence that BPA is an ovarian and uterine toxicant.

The determination was based on many, many, many research articles published in the scientific literature. I reviewed the studies that were identified in the Peretz article as supporting the toxic endpoints identified. Briefly, this table lists the experimental animal studies that were cited as providing strong evidence for ovarian and uterine toxicity of BPA.

I don't expect you to be able to read this actually, but let me walk you through the sort of design of this -- the major points. The first column on the left is a list of the primary authors for the studies that I

reviewed -- the animal studies that I reviewed by first author and publication year. Across the top are criteria that are applied to toxicology studies to ensure that the studies were conducted scientifically, according to generally accepted practice.

These are basically the DART Identification Committee's criteria for listing a chemical as a female reproductive toxicant.

So you can see Y's in green boxes and N's in purple boxes up there. The Y's in green boxes indicate that the study meets a particular criterion. I guess I forgot to mention that -- okay, I crossed the top of the criterion. So the Ys indicate that the particular study meets the criterion. The N's in the no box -- or the N's mean no that the study does not meet the criterion.

There are some U's, and they're in gray boxes. And the U's indicate uncertainty about the criterion, for example, whether the appropriate exposure timing was used to relate to human exposures. For example, there are some NA, not applicable, gray boxes also. And those are under whether litter effects were controlled. And in that case, it's usually because the effect was in the maternal animal and the litters weren't studied.

So now that you're oriented, the table shows my overall scientific judgment about the scientific evidence

supporting the listing of BPA under Prop. 65 for those studies.

I just want to talk about a couple of the columns. The first column is was the study design relevant to female reproductive toxicity? And you can see the most studies listed were. A few studies I indicated were not, because perhaps the effect evaluated was in male offspring rather than female.

The second column in, was the appropriate number of animals per dose used? And, in many, many cases, the number of animals per dose group was fewer than six. And so, many of these studies just did not have an adequate number of animals to identify a statistically significant result. The third column in, was the route of administration in the study appropriate? And for the neonatal -- for the neonatal exposures, I did consider subcutaneous exposure appropriate based on the literature, but otherwise injection exposures are not considered relevant to human exposures. And so many of these exposure routes in these studies were not relevant to humans.

So overall, there are a number of criteria that are just not met by a lot of these research studies. And that's what these studies are. They're research rather than toxicology studies.

So basically, this table does not show the outcome of the studies listed, but just how the studies match up with the criteria for listing, and the weight of the new experimental evidence between 2009 and 2013 does not meet the DART criteria for listing under Prop. 65.

--o0o--

DR. MILESON: I have a similar table of the epidemiology studies that were listed in the Peretz paper as supporting the uterine and ovarian toxicity. And the same organization holds for this table the first authors and the years of publication are in the first column. The listing criteria basically, or the scientific criteria are across the top, and green Y's indicate that the criteria were met, purple noes indicate that they were not.

And one thing that I do just want to mention is that many of these studies were conducted on IVF, in vitro fertilization, subjects and that to me caused a level of bias in selection.

So this table shows my scientific judgment about the epidemiology studies. And the weight of the new epidemiological evidence does not meet the DART criteria for listing under Prop. 65.

Thank you.

CHAIRPERSON GOLD: Thank you. Does that complete the presentation?

MR. LANDFAIR: That does complete our presentations. Thank you very much.

CHAIRPERSON GOLD: Thank you. So does the panel have questions for either the ACC or the ACMI presentations?

You had one. Go ahead.

COMMITTEE MEMBER PESSAH: Actually, I have questions on the PK opinions that were expressed. And so do you think that steady state levels of BPA, given the short half-lives, reflect possible peak levels following exposures especially during the critical periods of development -- gestational develop?

DR. HENTGES: So repeat again the question, make sure I got it? Thank you.

COMMITTEE MEMBER PESSAH: Yeah. You stated that there was first-pass elimination and very short half-life. The question I have is during pregnancy, what are the peak levels? Are you sure that they're not well above what you stated?

DR. HENTGES: Two points that I'll make on that. One is that based on everything we know about human exposure and pharmacokinetics, the levels of parent BPA, free BPA, in blood should be below current levels of detection, should be in the picomolar range not even close to nanomolar.

1 And the time profile has been analyzed in a study
2 published by FDA researchers. They've also develop a PBPK
3 model that they've applied. And so what they've done is
4 they've modeled what happens over, let's say, the course
5 of a day with, you know, BPA comes in through the diet, as
6 you point out, it has a short half-life. So things aren't
7 necessarily exactly the same at every time point.
8 And so I think if you look at that, the bottom
9 line is yes the levels would be below levels that should
10 cause any estrogenic effect. I don't know if I explained
11 that very well, but I could show you the papers.
12 COMMITTEE MEMBER PESSAH: So you're saying that
13 the free BPA levels in the blood would be below detection
14 levels or below -- certainly below the EPA levels, but
15 those in the urine would be for free BPA would be above
16 those levels?
17 DR. HENTGES: Not free BPA. In urine what you
18 find is the conjugate, the metabolites. That's what's
19 actually excreted. And I mentioned a study published just
20 a couple weeks ago from Johns Hopkins university. Even at
21 three days after birth, everything that came out in urine
22 was in the form of a conjugate. No free BPA at all was
23 found in urine.
24 And the reason urine is a little easier to
25 analyze is because BPA essentially concentrates in urine.

So it's -- I've seen estimates of maybe 30 to 100 times more concentrated as it comes out in urine compared to what it would be in blood. So it's a lot easier to measure, because the levels that you would expect to find are higher.

COMMITTEE MEMBER PESSAH: Right, but are you familiar with the Merritt study out of Columbia? They measured BPA in pregnant women in the urine, and what the levels were relative to total BPA, the ratio?

DR. HENTGES: I don't recall that study off the top of my head, no.

CHAIRPERSON GOLD: Other questions from the panel for ACC or ACMI?

Okay. Hearing none. We will go now to the individual public speakers. We hear -- and each of these will have five minutes. So Robert Chadwick from the Can Manufacturers Institute.

MR. CHADWICK: Hello. I'm Robert Chadwick, from the Can Manufacturers Institute. The Can Manufacturers Institute appreciates the opportunity to submit opposing written comments and brief testimony today before the DART Committee.

CMI is the national trade association of the metal can manufacturing industry and its suppliers in the United States. CMI member companies domestically produce

1 approximately 120 billion food and beverage cans annually,
2 and have more plants and more employees in California than
3 in any other state. Our members are committed to our role
4 in providing safe and nutritious foods and beverages to
5 consumers.
6 CMI written comments address the studies
7 currently under review by this Committee. Our testimony
8 today is about the safety of metal packaging and why BPA
9 is an important issue to the can manufacturing industry
10 and its customers, and reminds the Committee that your
11 actions today have real consequences.
12 And I guess with that comment, I trust the panel
13 will have no trouble faithfully executing their duties as
14 panel members -- or Committee members.
15 Around the world, food safety regulators -- or
16 food safety regulatory agencies have repeatedly concluded
17 that current dietary exposures to BPA do not pose
18 reproductive or developmental health risks. And I've been
19 advised that the Panel members have copies of this -- of
20 this testimony and there is a table attached to that.
21 Globally, most cans produced today use high
22 molecular weight BPA-based epoxy resin coatings, which
23 contain small amounts of residual BPA. These coatings in
24 metal cans preserve the container's integrity protecting
25 against microbial contaminants, and maintaining the food's

nutritional value.

The U.S. Center for Disease Control and the Food and Drug Administration estimates that each year roughly 128,000 Americans are hospitalized and 3,000 die of foodborne illnesses.

There has not been a single incident of foodborne illness from the failure of a metal can in over 30 years. Metal cans are not just packaging. The canning process commercial -- produces commercially sterile shelf-stable food. That means no E. coli, no listeria, no salmonella without any preservatives.

A Prop. 65 listing for BPA will discourage families from eating canned food, which could limit healthy and affordable food choices for children and adults. Canned foods make up about 17 percent of the American diet, and offer the lowest cost, most efficient means of delivering fruits and vegetables to the U.S. population helping meet USDA fruit and vegetable intake goals for Americans.

We believe the weight of scientific evidence does not support a BPA listing and we urge the Committee to oppose and not scare Californians from eating safe, economical choices like canned food and beverages.

Thank you again for the opportunity to provide testimony today and I'm happy to answer any questions.

CHAIRPERSON GOLD: Thank you. Any questions for Mr. Chadwick?

Dr. Luderer.

COMMITTEE MEMBER LUDERER: You described the current linings that are used in metal cans. Is this a -- and you mentioned the polymers of bisphenol A that are -- that form the lining. Has the can association done studies measuring the migration of any free bisphenol A into the foods in the cans in those with that type of lining?

MR. CHADWICK: There's quite a bit of published information available, studies that have been conducted from market surveys, where organizations have gone out into the marketplace, purchased materials off the shelf, and then conducted analyses on the food products themselves.

CHAIRPERSON GOLD: Dr. Luderer, do you have something else?

COMMITTEE MEMBER LUDERER: No, thank you.

CHAIRPERSON GOLD: Dr. Plopper.

COMMITTEE MEMBER PLOPPER: If there's these studies out here, we weren't provided these. So what are the levels that are in these food products, and does it vary by whether their lip -- they contain high levels of lipids or low levels of lipids, or they have ethanol in

them?

MR. CHADWICK: There's quite a bit of variability. One thing that's not readily apparent from the products is how complex and diverse the specifications and the materials are with the particular container and the particular food product.

We talk about epoxy coatings and epoxy resin coatings. There are well over 100 different, you know, types of epoxy coatings. So you'll have that a part of the variability. The food products comes into play. There isn't -- there isn't a specific trend relative to fatty foods versus aqueous foods. The variability is much more dependent upon the specific coating formulation and then very importantly the thermal process that's applied to sterilized the food product.

COMMITTEE MEMBER PLOPPER: You still haven't answered my question. I used to work with epoxy resins, so I understand all this.

MR. CHADWICK: Okay. Terrific.

COMMITTEE MEMBER PLOPPER: What I want to know is what ends up in the can? Maybe we need to see some of these studies. I mean, are we talking micrograms per ml, or milligrams per ml, or nanograms per ml?

MR. CHADWICK: It's micrograms per liter. That's our terminology, ppb. And depending upon the

specifications, you'll have -- you'll have a number of systems that are in the single digit ppb levels in the food product. You'll have others. There's another major category where you'll have averages in the, you know, maybe 35 to 70 ppb. And then there are other types of materials where you'll have higher levels, anywhere from 100 to 250 ppb.

And those are averages. There's a high degree of variability, because the BPA present is not intended to be there. It's just a residual from the manufacturing process.

CHIEF COUNSEL MONAHAN CUMMINGS: Can I just -- I apologize for interrupting. This is Carol Monahan Cummings. Were there any other questions for this witness?

Okay. I just -- I wanted to just --

CHAIRPERSON GOLD: Thank you.

MR. CHADWICK: Thank you.

CHIEF COUNSEL MONAHAN CUMMINGS: -- just briefly mention, especially for the newer members, that to -- as I mentioned in my earlier comments before we started, the process here that I know it's difficult to do, because it's not -- the Prop. 65 is kind of an unusual law, but the question before the Committee is not about whether or not the current human exposures to BPA are sufficiently

high to be of concern. So I understand there's been a lot of discussion about the -- it's totally fine for you to think about epi studies obviously, if there's Epi studies and there's blood levels and various things like that.

But the -- whether or not the current exposures, for example, Dr. Plopper, from migration from the epoxy to the food is, you know, at any level in particular, isn't a question that would inform the Committee about whether or not the scientific evidence shows that the chemical causes a particular effect.

So if you have questions about that standard, I know that a number of people have brought up the question what clearly shown means. And again, it is a scientific judgment call on your part. You do have guidance materials that were developed by your Committee several years ago. It's not a legal standard, and you don't have to determine today whether or not the listing will have any effect on any product or what kinds of exposures humans might have now or in the future. I hope that helps.

COMMITTEE MEMBER PLOPPER: Okay. I need to follow up with that, because we just heard a series of presentations that denied that some of the more strongly scientific studies were not relevant because of various conditions as exposure, because they don't represent what

1 happens in humans.
2 So that's my difficulty with this is that if
3 we're going to disregard those, and we're looking at them
4 strictly as scientific studies that are not necessarily
5 related to one paradigm of how humans are exposed, and
6 that's my concern is because if my -- I'm hearing what
7 you're saying is that we disregard these other issues and
8 look strictly at the science directly with -- and not
9 related to --
10 CHIEF COUNSEL MONAHAN CUMMINGS: Yes. What I'm
11 trying to explain is that that is true. You need to look
12 at the scientific evidence that's presented here. I'm far
13 from being a scientist, but this particular Committee it
14 is -- the charge is somewhat unusual, because of the way
15 that the statute was drafted. We don't have regulatory
16 criteria, other than what the actual language out of the
17 statute that says that it has to be clearly shown by
18 scientifically valid testing, according to generally
19 accepted principles to cause reproductive toxicity. And
20 that's why the Committee in the past developed the
21 criteria that you have as guidance.
22 It is not a straightjacket. It is definitely not
23 meant that way. It was kind of a help to kind of parse
24 through the evidence. And so there -- you shouldn't
25 discount the fact that there are human studies. What I'm

saying is that the current exposures to humans right now is not a concern for this Committee. It's not something that's part of your criteria, and it is something that would be addressed later in the Prop. 65 process when there's determinations about levels of exposure that require warning for example. And that's something that our Office does. And you all, as peer reviewers, would review that information at that point. Does that help?

COMMITTEE MEMBER PLOPPER: I think so.

CHAIRPERSON GOLD: Okay. Thank you. Are there any other questions from the panel for this last testimony?

If not, we'll proceed with the remainder of the public comments. So next is John Rose from NAMPA, five minutes.

MR. ROSE: Thank you for the opportunity to talk here today. As I think you were just told, although exposures are not relevant here, I think it is important to look at studies and understand if the relevant doses of those studies are even in relative orders of magnitude of what humans are actually exposed to in the blood stream. Although, like you said, as you were just told, that's not necessarily your purview today, but it's important to look at it, and under understand that every chemical, at some level, is going to be harmful.

So following that criteria, it would reach a point where everything would have to be listed. So there has to be some sort of threshold where it has to be at least a relevant dose within a couple order of magnitude.

So as we know, humans are exposed almost exclusively from BPA by oral exposure. Recent pharmacokinetic studies have shown that free BPA in the blood stream is rapidly metabolized at greater than 99 percent to the non-biologically active bisphenol A glucuronide. And as Dr. Hentges mentioned, there has been a lot of recent research that has looked at the contamination level -- contamination issue, and that a lot of studies that have been published actually have significantly higher levels than now, what we're understanding would actually be in the blood stream.

In fact, it's sort of standard practice now that you have to identify and list not only the free BPA but the metabolized BPA, so that you could look at those ratios. And if you're seeing a ratio far off from 99 percent of the metabolized level from the free BPA, it's almost certainly coming from contamination issues. So many of the studies that go back more than just a couple of years before this issue was identified sometimes identify much, much higher levels of free BPA in the blood stream than actually could ever occur.

And in our written testimony, we did -- the main point of that was to look at the review article that we're discussing today. And what we did was basically look at all those studies and highlight for you the opinions of those studies by USFDA and EFSA and almost exclusively those studies were dismissed as not relevant for hazard identification or risk assessment.

So as we go forward with this -- your discussions today, it's important to note that a decision to list this would be the first government panel to do so to make a statement about the safety of BPA, which would be quite inconsistent with many of the other recent assessments by USFDA, European Food Safety Authority, and many other panels over the last few years.

Thank you.

CHAIRPERSON GOLD: Thank you. Do the panel members have any questions?

Dr. Pessah.

COMMITTEE MEMBER PESSAH: Are you aware at the rate of UDP-glucuronyl transferase polymorphisms in the human population?

MR. ROSE: Say it one more time?

COMMITTEE MEMBER PESSAH: You mentioned that glucuronidation is a major pathway to essentially neutralize BPA's estrogenic effects or endocrine

disrupting effects. That's a highly polymorphic gene, where there's a substantial number of individuals in the population that are never accounted for in epidemiological studies, at least not ones that I've seen, which impair glucuronidation. Have you considered that in your analysis?

MR. ROSE: It hasn't been considered but the number of studies I can't say it hasn't been considered. I have not considered it. But the number of studies that have looked at the level of glucuronidation would not suggest that that's an issue just based on the statistics of -- as you said, there's a high -- significant number of people that hasn't been shown in those, in that research.

COMMITTEE MEMBER PESSAH: Well, that's never been actually controlled for in any studies.

MR. ROSE: Not that I'm aware of, but I'm not certain about that.

CHAIRPERSON GOLD: Any other panel members have questions?

Okay. The next speaker I can't tell the first name, so Mr. or Ms. Rodriguez from Center for Environmental Health. Mister.

MR. RODRIGUEZ: Hi. I'm Brian Rodriguez. I'm a current graduate student at the UC Berkeley Environmental Health Science Department. Today, I'm representing the

Center for Environmental Health in Oakland. And on behalf of our 5,000 California supporters, I want to say thank you for letting me talk to everyone.

I want to emphasize that the Center for Environmental Health and its supporters fully support the listing of BPA as a female reproductive toxicant under Prop. 65. OEHHA scientists have done an expert compilation of the large number of studies relevant to this topic. Our scientific evaluation of these studies confirmed the criteria for a Prop. 65 listing.

We encourage you to do the same as we believe that you'll find that it is scientifically sound. Prop. 65 has an almost 30-year history of protecting California consumers. Just some of the few examples of over the last few decades include the removal of lead from candy, the removal of arsenic from playground equipment, and the removal of flame retardants from furniture and crib mattresses.

Your work in ensuring that -- your work in ensuring that -- sorry. Your work in ensuring that the current literature is backed in this Prop. 65 listing is critical, and I want to thank you for that.

Thanks.

CHAIRPERSON GOLD: Thank you. Are there any questions for this speaker from the panel?

Okay. Thank you very much.

So our next speaker is Gretchen Lee Salter.

MS. SALTER: Good afternoon. Thank you so much for allowing me to give comments. My name is Gretchen Lee Salter. It's good to be back. I worked on BPA in my capacity at the Breast Cancer Fund for many years. But I no longer work at the Breast Cancer Fund, and today I'm just here as a concerned citizen and mother of two young daughters. I have no conflict of interest here. I have paid my own way here, because I care deeply about this issue.

I am not a scientist, and I'm not going to talk about the science today, but I do want to talk about the implications of your actions today from the public's perspective. I consider myself to be a incredibly educated about BPA, especially about what products I can find it in. I have given talks about this subject to the public and educated other mothers about what to look for when trying to avoid BPA.

But even as an educated person, I cannot definitively say what has BPA in it and what does not. When I go to the store and look for canned beans or tomatoes or when I grab a receipt from a vendor, I have no idea if it contains BPA or not.

I know the studies, and I know what the studies

show that exposure to BPA, especially in utero, can lead to increase risk for later life harm and disease, including impacts on the female reproductive system. When I was pregnant a little over a year ago, I shrank back from accepting receipts and from eating canned food, because I had no idea if they contained BPA or not.

It would have been much easier to know, one way or another, whether these items contain BPA. Ask any mother to look at the number of studies showing an impact from BPA and whether or not she would want to know if her products contained this chemical, and her answer would be emphatically, yes.

California's Prop. 65 program has received a lot of criticism from those in industry saying that these warnings aren't helpful to the general public. I completely disagree. Information is power. Knowledge is power and they know that. I have seen the song and dance that industry puts on when it comes to BPA for over 10 years. Their intent is to obfuscate, confuse, and overwhelm so that no decision is made, and so that they can continue making billions of dollars every year making and using this chemical.

They are here because they have a financial interest to be here. I think it is important to mention here that the same considerations do not motivate me or

color it -- or color the members of the NGO community here today. As I have said, I have taken time away from my girls, paid for child care, and paid for my way up here personally, so that I can make these comments to you.

The NGO community making statements here today are here because they are concerned about the public welfare. They do not receive a bonus or an increase in share price if BPA is listed. They merely have the satisfaction of knowing that the public is that much more protected. As I can attest as a former member of the NGO community, you do not get rich for fighting -- by fighting for public welfare.

I have seen industry try to argue the science. The overwhelming evidence shows that BPA clearly causes reproductive harm. I have seen them try to argue the legal arguments. What does clearly shown mean? Does BPA meet the standard?

I am astounded at the time, effort, and money that they are taking to make sure that this chemical isn't listed. As an advocate it infuriated me, but as a mother it makes me sick. How can they look at the data you have received and not even have one ounce of concern? How can they stand there and advocate for the continued uninformed exposure of pregnant women when there are hundreds of studies showing -- staring them in the face about the

impacts associated with BPA exposure.

It is almost as though they think that your job is to ban the chemical. But we need to remember, this isn't about a ban. This isn't about real world exposures or not. This isn't about whether BPA has been shown to cause -- I'm sorry, this is about whether BPA has shown to cause reproductive toxicity. That is it. Does it meet the criteria set out before this panel for listing?

From the discussions and the presentations, I don't know how it is possible for the panel to come to any other conclusion than to answer yes. I pray my knowledge about BPA and how to avoid it has kept my girls safe from exposure.

I ask -- no, I beg for this Committee to follow the science, to do -- to do what is necessary to inform other mothers in the future by placing BPA on the Prop. 65 list.

Thank you.

CHAIRPERSON GOLD: Thank you. Are there any questions from the Committee?

No.

Thank you.

MS. SALTER: Thank you.

CHAIRPERSON GOLD: Next we have Emily Reuman from the Breast Cancer Fund.

MS. REUMAN: Hello. Thank you so much. My name is actually Emily Reuman, just to clarify. And I'm here representing the Breast Cancer Fund and its thousands of supporters in California. And on behalf of the Breast Cancer Fund, I just first off want to thank the panel for examining the science on BPA. We really appreciate the opportunity to speak publicly about this matter, and we are very encouraged that State scientists are taking such a careful look at this toxic, hormonally active chemical.

And I also want to thank so much to the staff for clarifying for all of us that questions about risk and exposure are not the questions this Committee are addressing today. Today, we're only concerned about whether or not the evidence before you demonstrates female reproductive toxicity. Either human or animal studies are sufficient for listing. And based on the presentations made today by OEHHA staff, those criteria have been more than met.

Founded in 1992, the Breast Cancer Fund works to prevent best cancer by eliminating our exposure to toxic chemicals and radiation linked to the disease. Our work to fulfill that mission brought bisphenol A to our attention in 2001 during the development of the first addition of our report, *State of the Evidence: The Connection Between the Environment and Breast Cancer*.

Early scientific studies identified BPA as an endocrine disrupting compound that altered development of the mammary gland animals alterations that increased the risk of mammary cancers later in life. After nearly 15 years of collaborative work, environmental health science and advocacy, we now recognize that BPA is linked, not only to breast cancer, but to alterations in the development of reproductive, metabolic, immune, and neurobehavioral systems in humans and animals.

And today, the body of evidence has grown significantly to include studies that show exposures to even extraordinarily low doses of BPA, particularly during prenatal development and early infancy are associated with a wide range of adverse health effects later in life.

Exposures that occur before birth are particularly troubling as the effects on developing fetuses are irreversible. The Breast Cancer Fund published a report in 2013 summarizing research to date on the health effects of prenatal BPA exposure, disrupted development, the dangers of prenatal BPA exposure.

This report documents the mounting evidence linking BPA exposure in the womb and soon after birth to health effects, including breast cancer, prostate cancer, metabolic changes, decreased fertility, early puberty, neurological problems, and immunological changes.

Significantly many of these studies document negative health effects from low dose BPA exposure. Most of the doses much lower than EPA safe dose.

The science is clear, BPA causes a wide range of developmental and reproductive effects. The materials that have been prepared by OEHHA staff demonstrate clear reproductive toxicity harm from BPA. In addition, I thank and I urge the Committee for closely examining flaws in studies presented by manufacturers of bisphenol A.

And while these interests claim that BPA does not cause harm or that the science is unclear, we ask the panel to recall that we have heard similar protestations before within the tobacco industry and the lead paint industry.

These industries wanted to continue using these products that scientists knew were harmful and therefore manufactured their own science to support their aims, causing unwarranted doubt, uncertainty, and inaction on the part of regulators that lead to the needless harm to Californians and the American public.

We must not allow industries that stand to gain financially from your decisions to continue to cloud this issue. The Breast Cancer Fund urges you to consider the evidence today that this chemical should be legally identified as a reproductive toxicant. And failing to do

so would knowingly put the public's health at risk.

Thank you so much for all of your good work.

CHAIRPERSON GOLD: Thank you.

Are there any questions from the panel for this witness?

No.

Thank you very much.

The next speaker is Bill Allayaud from Environmental Working Group. I'm sorry, if I mangled your name.

MR. ALLAYAUD: Hi. I'm Bill Allayaud with the Environmental Working Group here in Sacramento. We work on issues in environmental health, what you get exposed to in your food, your water, what you put on your skin.

Again, the question here is not whether you should ban a chemical or how to label it. We leave that to OEHHA, and they're doing a good job of revamping the Prop. 65. Labeling things is a tough job. It's here really to say is BPA toxic to the female reproductive system. The European Union is strengthening its reproductive toxicity categorization of BPA right now based on the weight-of-evidence approach.

The European Union's Committee for Risk Assessment, the RAC, which prepares the European Chemical's Authority's opinions of risk and -- of

substances to human health and the environment has adopted an opinion to strengthen the classification of BPA to Category 1B reproductive toxicant or one that is quote, "Presumed to produce an adverse effect on reproductive ability or capacity or on development in humans", unquote.

Listing BPA as a reproductive toxicant under Proposition 65 is in harmony with this recategorization by the EU. The RAC opinion over there was based on the weight-of-evidence assessment that showed clear evidence of adverse effects on sexual function and fertility in animals with a mode of action that is relevant to humans. This reaffirms that BPA meets the criteria for listing under Proposition 65.

The mode of action for disruption of the reproductive tract described in the opinion included a direct or indirect disruption of the HPG axis direct organ specific toxicity and BPA interaction with estrogen receptors. The opinion states quote, "Early BPA exposure during the period of brain sexual differentiation may exert indirect effects on reproductive tract tissue by altering the function of the HPG axis, an effect would become apparent after puberty", unquote.

While the EPA -- FDA has submitted comments against the listing, as the ACA has pointed out, the FDA does not make a sound case that BPA is not a reproductive

toxicant. In an FDA letter to your Committee, the agency stated that their assessment of BPA does not support its listing under Prop. 65. However, their evaluation focused on whether or not BPA used in food contact substances results in an unsafe level of exposure. This is different from the comprehensive weight-of-evidence evaluation of whether or not BPA has the ability to cause reproductive toxicity, an endpoint that has been clearly demonstrated in animal studies and supported by human data.

FDA also excluded most independent peer reviewed publications reporting reproductive toxicity from its formal hazard review process. The FDA did identify sperm testicular hormone-related parameters as hazard endpoints, which are reproductive endpoints. Developmental neurotoxicity was also identified as a hazard endpoint.

The FDA does, in fact, identify potential reproductive hazards in its review, which include female reproductive endpoints, such as follicle and oocyte development in ovary estrous cyclicity and effects on the HPG access and puberty onset.

However, the agency excludes from most -- excludes most independent peer-reviewed reports on BPA and reproductive toxicity from its hazard ID process for various reasons, such as statistical power sample size.

In addition, a 2009 paper by Myers et al.

strongly criticizes FDA for ignoring hundreds of independent academic peer-reviewed publications in their assessment of hazards associated with BPA largely because they were not good lab practices compliant. A weight-of-evidence approach does not exclude most academic peer-reviewed reports from the hazard identification risk assessment process.

The FDA argues against concern for BPA toxicity in people because humans metabolize BPA more efficiently than rodents. Conjugated BPA is considered inactive. However, a 2013 study reported that up to 90 percent of sublingually administered BPA was bioavailable. This indicates the potential for substantial systemic absorption of BPA from the oral mucosa, which invades -- evades detoxification by first-pass metabolism.

I'm running out of time, so I'll conclude by saying we think the evidence clearly supports to a listing and urge the DART committee to do so.

Thank you.

CHAIRPERSON GOLD: Thank you.

Any questions from the Committee for this?

Thank you very much.

The next speaker is Renée Sharp also from Environmental Working Group.

MS. SHARP: Tasha Stoiber is going to go first.

CHAIRPERSON GOLD: I can't hear you. I'm sorry.

MS. SHARP: Tasha Stoiber is going to go first.

CHAIRPERSON GOLD: Okay. Tasha Stoiber, is that right?

DR. STOIBER: That's fine.

CHAIRPERSON GOLD: Okay. You're both from Environmental Working Group and you would like to go first.

DR. STOIBER: Yes.

CHAIRPERSON GOLD: That's fine.

DR. STOIBER: Thank you for the discussion and the time to speak today. My name is Tasha Stoiber and I'm an environmental chemist and senior scientist at the Environmental Working Group. I have no conflicts of interest with anything discussed today. EWG is a national nonprofit research and advocacy organization and they paid for my travel to be here today.

I would like to comment that the weight of scientific evidence shows that BPA meets the criteria for listing as a female reproductive toxicant. Over the last decade, new research on reproductive toxicity has become available that provides strong scientific evidence that BPA is a female reproductive toxicant.

Some of the recent data is summarized in the Peretz et al. article that has been submitted to the

Committee and discussed in depth today. And based on the current weight of evidence, the authors conclude that BPA is a reproductive toxicant, a position that Environmental Working Group strongly supports.

This scientific conclusion was based on multiple lines of evidence in in vitro experiments, in vivo animal models, and associations in humans. In addition, adverse effects have been demonstrated for multiple reproductive endpoints in female animals and people. The findings from original research that are reviewed by Peretz et al. clearly show that BPA meets the criteria for listing.

Specifically, gestational exposure to BPA can effect egg production by disrupting the onset of meiosis in the ovary. This has been observed in rodents and reconfirmed in primates at BPA levels that have been observed in humans. Follicular defects have also been reported in rodents, sheep and primates.

Recent research has also shown that BPA exposure may affect the uterus and endometrium. Gestational exposure produced changes in uterine morphology and adult rodents and hens. A case control study in women showed an association between BPA concentrations in serum and endometriosis.

Experimental studies in rodents and in vitro studies support the premise that BPA adversely affects the

uterus. In humans, some studies suggest that increased body burden of BPA is associated with decreased fertilized eggs in women undergoing IVF treatment. A recent study found urinary BPA concentration adversely affected implantation outcomes in women and several animal studies supported this effect from both exposed female and male rodents. Notably, even when only the male rodents were exposed and not the females, implantation was also adversely affected.

Reproductive effects reported in animal studies across species and the associations between BPA and adverse reproductive outcomes in women provide strong evidence that BPA is a reproductive toxicant. There is also significant support BPA as acting as a reproductive toxicant in men. These findings are supported by mechanistic data, including studies on hormone receptor interaction and gene expression.

It's also important to consider research on especially sensitive populations, including the fetus and newborns. Research demonstrates that exposures in the womb and neonatally produce adverse reproductive outcomes. The pathway that detoxifies BPA is not fully developed in the fetus or young infants. Biomonitoring studies reviewed by Vandenberg et al., 2010, showed unconjugated BPA present in pregnant women, umbilical cord blood and

serum, placental tissue, amniotic fluid, and breast milk.

This poses a unique risk to both in utero and after birth. As evidenced in relevant animal studies, such exposure may result in reproductive effects later in life.

Again, considering the numerous scientific studies that have been examined, we strongly support BPA listing as a female reproductive toxicant as the criteria have been met.

Thank you.

CHAIRPERSON GOLD: Thank you.

Do the Committee members have any questions for this?

Thank you.

DR. STOIBER: Thank you.

CHAIRPERSON GOLD: So now Renée Sharp.

MS. SHARP: Thank you. Thank you for allowing us to switch places.

So my name is Renée Sharp. I'm the Research Director for Environmental Working Group, a nonprofit research and advocacy organization. And I have no financial interest in the outcome of this hearing today, and I want to make several points.

First, in contrast to what was stated earlier by ACC and ACMI, there are at least five studies showing free

BPA in urine, all which were reviewed in a paper published by Vandenberg et al. in Environmental Health Perspectives in 2010 titled, Urinary Circulating and Tissue Biomonitoring Studies Indicate Widespread Exposure to bisphenol A. So I just wanted to clarify that.

Second, I want to reiterate what Carol Monahan from OEHHA stated earlier about how the Committee's task is not to consider exposure. However, since there were questions about how much BPA leached from cans, I thought I would just note that the BPA that -- that BPA has been shown to leach from cans at levels of up to 1000 Micrograms per kilogram, which is not a small amount.

Third, I want to note that there are over 90 epidemiology studies suggesting harm from exposures and many hundreds of animal studies showing harm from low doses.

Fourth, I also want to address the Delclos et al. study, which was conducted under the guise of FDA, as you all know, which the chemical industry has discussed at length in their comments earlier today.

The fact is that this study has serious problems, unfortunately. Notably, the control animals were accidentally exposed to BPA. And the control animals actually had BPA exposure equivalent to the low dose groups. Therefore, the study's conclusion about low dose

effects is invalid. And this is not just my opinion. Pat Hunt et al. published a paper in Toxicological Sciences that concluded that quote, "Contamination and negative controls renders this control group useless for assessing low dose effects".

It's also notable that, nevertheless, EFSA actually looked at the study and concluded there were actually mammary effects from -- that were shown in the study.

Fifth, I want to underscore again the criteria for listing. I do this because over the past 14 years that I have been coming here and testifying, I have seen previous DARTIC committees routinely seem to get confused about what the task is that is set before them.

So please indulge me as I review this again. I know you've heard it a lot. But after 14 years, I feel like it's actually my duty to do this, because somehow it seems to get confusing.

So once again your task is if there's clear evidence for female reproductive toxicity in animals or humans, you must vote to list. And if there's only one endpoint clearly showing female reproductive toxicity, you must vote to list. So that is what the law says, and I would say that all evidence presented here clearly points to the necessity for listing.

And finally, I just want to make one point regarding the ACC's request to have a couple of the members of the DARTIC committee recuse themselves from the deliberation. Just in thinking about future precedent, we, at the EWG, are concerned about the precedent that independent scientists who were appointed to the Committee because of their expertise, because of their scientific work would be prompted to recuse themselves because of their work. They don't have a financial interest in this. They are intellectually unbiased. So we just believe that that is just not -- not a good precedent to have and just wanted to make that final point before I urge you to list. Thank you.

CHAIRPERSON GOLD: Thank you. Any questions for Renée Sharp?

No, thank you.

Okay. Our last person to speak is Rebecca Sutton from San Francisco Estuary Institute.

DR. SUTTON: Everyone can hear me? Oh, yes.

All right. Thanks for the opportunity to speak. My name is Dr. Rebecca Sutton. I've a Ph.D. in environmental chemistry, and I'm a senior scientist with San Francisco Estuary Institute, where I lead focus areas in emergent contaminants, bisphenol A would be one of those, and green chemistry. I'm also a member of the

Green Ribbon Science Panel, which is a Department of Toxic Substances Control expert panel. We're there to help the Department implement its Safer Consumer Products Regulations, but I'm not here representing that Panel or DTSC. I'm here representing SFEI, San Francisco Estuary Institute, and I don't have any conflicts of interest.

So I want to introduce you to SFEI very briefly so you can see I have a bit of a unique voice. Kind of great that I'm coming in last here. We are a research institute. Our goal is basically to develop the science to fill data gaps for stakeholders, policymakers who are considering different management actions when it comes in particular to pollution or ecosystem health.

So we don't, for example, take positions on bills or legislation. We're here as scientific resources typically for local agencies, regional agencies, State agencies and sometimes other stakeholders. So that's just to introduce my organization as a little bit different than all the previous speakers.

We've been following bisphenol A for a number of years how. We're concerned about it as a bay pollutant. Now, some of the research that I follow -- I follow also the human health literature, because we're also concerned about human health, but I also look at a -- perhaps a broader range of animal subjects than you all might. Just

as an aside, there are concerns in the non-human realm and the non-mammalian realm, regardless. The literature that you've reviewed and that I've reviewed regarding bisphenol A would seem to indicate that we've got a definite weight of evidence here, substantial literature indicating this chemical is toxic to female reproduction.

So we have a lot of animal studies. We have very suggestive human epidemiological data, and we're seeing the Salian in vitro work that's starting to pinpoint some potential mechanistic pathways for how this is occurring. So we see this chemical as toxic to female reproduction based on the current state of science and the weight of the evidence. It's guiding our current work. We have some active research again on fish not humans, in terms of endocrine disruption, gene expression, and developmental effects.

And so since I'd already done this sort of research and review internally, I wanted to bring it forward to you guys as a different set of stakeholders and decision-makers, because that's basically our role is we're trying to bring that science to the various decision makers and then turn it over to them and let them make the decision.

I would say also this is a bit of personal issue for me. I am a mom. I have an 18-month old. And just on

a personal level, it did take me a really long time to get pregnant for unknown reasons. I didn't have to go the IVF route like some of the folks we read about these studies. But I do wonder. I don't have a family history of this, and I certainly wonder whether chemical exposures could have played a role.

Again, exposure isn't the question, this is a toxicology matter, and exposure would be something we handle in a different framework, not -- well, different meeting, not this one. But I just wanted to bring that up as a personal note.

Any questions?

CHAIRPERSON GOLD: Thank you, Dr. Sutton.

Any questions from the panel?

Thank you very much.

So I think what we'll do is take a break -- brief break.

MR. LANDFAIR: Dr. Gold, I'm not going to speak, but I have paper copies of our slides. I'd like to give one to the clerk for the record and I'd like to distribute them to the panel if I may?

CHAIRPERSON GOLD: Thank you. Oh, oh. Okay. It took me a minute to understand it. I guess we do have one more speaker, Dr. Veena Singla. I didn't realize this was a separate presentation. So we'll take five minutes for

this presentation, and then we will take a five to ten minute break.

DR. SINGLA: Thank you. Veena Singla with the Natural Resources Defense Council. And I'm a staff scientist in the Health and Environment Program there. And NRDC paid for me to be here today.

And I wanted to just clarify a couple of points made earlier on the determinations made on the hazards of BPA by EFSA and the European Chemicals Agency. And in EFSA's hazard assessment, they did find, based on their evaluation of the weight of the evidence, that BPA was likely to have effects on the mammary gland. And as one of the previous commenters mentioned the European Chemicals Agency also found that BPA was a presumed reproductive toxicant in their latest evaluation last year.

And as a number of commenters have noted, I wanted to speak to the importance of the question before the panel. One of my favorite quotes from Albert Einstein is, it goes something like he says, you know, if I was trying to solve the most important problem in the world, if I had one hour, I would spend 55 minutes figuring out what the right question is to ask, and then five minutes solving the problem.

So here the question before you is simply, is

there sufficient evidence from studies that BPA is a reproductive toxicant, human or animal studies? And that's the simple question to answer, not questions about what is a safe level or what is the NOEL or is the current level in food safe, but simply is BPA a reproductive toxicant?

Thank you.

CHAIRPERSON GOLD: Thank you.

Are there any questions for Ms. Singla?

Okay. So I think we should come back at 3:20, does that sound good? Let's aim for 3:20 just for a get up and stretch kind of a break.

(Off record: 3:15 PM)

(Thereupon a recess was taken.)

(On record: 3:23 PM)

CHAIRPERSON GOLD: Okay. Are we ready to resume? Is everybody here?

Oh, yeah. I couldn't see you over there.

We don't have OEHHA staff. Okay. At this point in the agenda, the next thing is for the Committee to discuss everything that we've heard today, and eventually see if we're ready to take a vote to list or not. But at this time, I'm opening it up for the Committee for a discussion.

So who would like to start?

Dr. Luderer.

COMMITTEE MEMBER LUDERER: Sure. I just have a few comments to make. And one of the things that I think is really very important is not -- that it's important for us to really assess all of the studies and not to dismiss scientific studies because they examined different endpoints in many cases than the traditional regulatory studies. They may not have been done according to GLP. However, I think it's important for us to examine all the studies and look at them as a whole as a body of scientific literature and come to a conclusion based on that, rather than excluding them from consideration.

I also wanted to -- I appreciate Carol Monahan Cummings making the point that we are not here today to determine a safe level of exposure, but really to make an assessment about whether this is a female reproductive toxicant.

I think it is though important to address the issue of whether the non-oral routes of exposure are relevant to humans or not. There have been several recent papers, I'm thinking particularly of papers by Herman et al. and Gayrard et al. that showed significant dermal and sublingual absorption of bisphenol A.

And there have also been several studies showing that subcutaneous and oral exposures result in similar

1 serum levels in neonatal rodents. In addition,
2 concentrations of free bisphenol A in humans that have
3 been measured in serum, some of the more recent studies
4 have measured the serum concentrations. And these were
5 done by the CDC labs, of free bisphenol A in rodents with
6 subcutaneously implanted mini-pumps as the route of
7 exposure, and shown that the serum concentrations of
8 unconjugated or free BPA were similar -- or were in the
9 range of what has been reported in the human population.
10 So I think that those routes of exposure in those studies
11 cannot be dismissed on the basis of that.
12 And finally, I think it's important to note that
13 biomonitoring studies have repeatedly shown that BPA is
14 measured in nearly all humans. And therefore, people are
15 repeatedly exposed multiple times a day, given the short
16 half-life of bisphenol A.
17 So thank you.
18 CHAIRPERSON GOLD: Thank you.
19 Dr. Pessah.
20 COMMITTEE MEMBER PESSAH: Thank you. I want to
21 point out that my expertise is not female reproductive
22 toxicology. And so when I approached the literature, I
23 approached it from a very sort of neutral perspective with
24 respect to looking at the data and trying to decide
25 whether or not there was weight of evidence.

1 Clearly, BPA is a pervasive exposure issue, that
2 it is everywhere and humans are being exposed. The
3 question of levels of exposure is a good one, but that
4 argument doesn't incorporate the fact that we're all
5 different, and that when the major elimination route is
6 glucuronidation and the polymorphic rate of glucuronyl
7 transferases are such that different individuals have
8 different abilities to glucuronidate. And as we heard
9 someone say, that newborns don't develop their
10 glucuronidation potential until a couple years out, that
11 the potential harm is there from exposure.
12 Now, I viewed the literature especially, the
13 animal literature, which I was asked to review as being
14 variable. But one thing really came out that converged on
15 potential harm, and that is that the exposures, whether
16 they're bracketed above or below the EPA limit,
17 essentially caused shifts in gene expression. It's not
18 the usual D.A.B.T. kind of outcome. It is not the
19 classical 19th or 20th century toxicological verdict. It
20 is the new verdict.
21 And the fact if you then speed forward and say
22 how do those early changes in gene expression influence
23 outcomes in future generations, and you look at the PNAS
24 article that was published last year from the Columbia
25 Group and find that WNT pathways are disregulated in the

offspring at concentrations below those of the EPA levels. And those were, in fact, very solid studies that incorporated not good lab practices, but good scientific practices, in terms of the N, in terms of making sure that the mice were not exposed before the exposure.

That one has to think about if there is a five percent exposure rate with the potential of causing harm, what's the outcome to the kids that are produced down the line from these individuals that have epigenetic marks changed, especially maternally imprinted genes.

So with that, I would ask that we start to think about how genetic changes, not in the form of causing mutations, but causing changes in transcription that persist, influence potential negative and harmful outcomes.

CHAIRPERSON GOLD: Thank you.

Anyone else?

Other comments?

Dr. Kim.

COMMITTEE MEMBER AUYEUNG-KIM: So in echoing Dr. Luderer's and Dr. Pessah's comments that, you know, I, myself, you know, studied glucuronidation when I was in graduate school, as well as, you know, been in the environmental field, as well as the pharmaceutical field. And so essentially, I took the weight-of-evidence approach

as well in looking at the data. And that a large body, although, you know, individually the studies may not indicate that there is, you know, a reproductive -- female reproductive effect due to some of the limitations of the study, is that overall when they all point in the similar direction that that is an important factor to take into consideration.

CHAIRPERSON GOLD: Thank you.

Dr. Carmichael, Dr. Baskin, Dr. Plopper?

Dr. Plopper.

COMMITTEE MEMBER PLOPPER: I've already been told that this is not on our table, but when I evaluate these studies, I had the problem that the paradigm under which FDA judged those studies is that the only exposure route is material that gets into the digestive system from the small intestine to the large intestine, and that all of those materials are carried to the liver via the hepatic portal system, all right?

Well, first of all, there's no mention of the lymphatic clearance from the gastrointestinal system. And as you -- those of you that are aware, they're called lacteals, almost all of the fats that are digested fats end up being carried by the lacteals into the thoracic duct and into the left brachial venus vein, okay? So the assumption is here that glucuronidation is highly active

in the liver, and it's also in the mucosa of the intestine.

Well, unless -- I've only worked with six different metabolically activated toxicants in my career, but none of them successfully had 90 percent clearance on first pass, okay?

And the paradigm here is it's a hundred percent. Well, let me point out that if you use four -- the standard dose now is 100 micrograms per kilogram, which translates into 100 gram rat as about 1000 nanograms, if 0.1 percent of that is not metabolized, you will have nanogram quantities in the blood stream. That is what's been observed in humans.

So I have a problem with that paradigm as exposure. And being an exposure person for my career, we know that there are three barriers between the organism and the environment that have their interactions, and that's gastrointestinal track, the integumentary system, and the respiratory system. It is without doubt, in my experience, that a small molecular weight compound like BPA, which is lipid soluble, is going to be rapidly passed through the barriers of various aspects of the skin, as well as in the oral cavity.

And having worked with monkeys for over 40 years, I know that monkeys don't chug their food. They don't

bolt their food. We don't bolt our food. They're -- the oral cavity is a very active site for absorption of pharmaceutical chemicals.

I will point out that nicotine a non-lipid soluble compound is used pharmacologically by everybody that chews tobacco. Okay. That's one thing and those of you that know someone that has cardiovascular disease, and I happen to be one of those, knows that one little nitroglycerin tablet under your tongue works effectively within 30 seconds to a minute.

So to assume that the only reliable studies that can be done to evaluate reproductive toxicity in animals or people is -- has to go through the digestive tract and only through a limited part of it, and that the liver, by some miracle, takes this particular compound and does 100 percent biotransformation, I find biologically unacceptable, and I don't believe there's any literature to say that this is true.

And when you look at the literature we were provided, and we looked up -- it's like Isaac and others have said, it doesn't take much material in the oral cavity, of three or four very recent studies, to bring the level of unconjugated BPA up into the nanogram per ML quantities. There is a vast literature we've discussed today that says those levels are biologically active.

Now, why would they not be biologically active in humans?

That's -- this is my concern is that if we are going to look at this -- if we're not going to be able to consider the exposure issues, then we have to ask ourselves why are these studies not appropriate that don't use one paradigm for their evaluation. If the idea was to set what is the exposure standard, which we're not doing, correct --

CHIEF COUNSEL MONAHAN CUMMINGS: Yep.

COMMITTEE MEMBER PLOPPER: -- then that's a whole nother issue. This is not the issue. This issue is the exposure is by the circulation, and there is no question that the reproductive tract of females in every species has been looked at that is biologically active at concentrations found in people.

And I will also point out there's this idea of how long does it stay is not relevant, because some of the shortest exposures of other compounds for the shortest period of time at a very high dose that then disappears may actually be more toxic than it is of if it's exposed at half that level continuously. It's called the development of tolerance.

And as Isaac said, glucuronidation is a genetically variable thing in people. It's also a site specific variation in people, so we don't really know what

this is having an effect on. And maybe I've said too much.

So I think that if we're going to look at the biological significance of this, then we have to disregard most of the paradigms that have been used to exclude specific studies. And that's all I'll say.

Thank you.

CHAIRPERSON GOLD: Thank you.

Anymore comments?

Does this mean we're ready to consider a vote?

Everybody ready?

Ready?

Okay. I have the language, right?

We're ready?

Yes. Okay. So the question before us is has bisphenol A, BPA, been clearly shown through scientifically valid testing, according to generally accepted principles to cause female reproductive toxicity?

So I'm going to request those of you who believe yes to raise your hand.

(Hands raised.)

CHAIRPERSON GOLD: Seven. That would be zero no votes, correct, and zero abstentions.

So the results is we have seven voting in favor, correct?

Okay. Thank you.

I believe our mission is somewhat done, except that we are going to hear about staff updates now, correct.

CHIEF COUNSEL MONAHAN CUMMINGS: Can I get the slide up, Esther?

(Thereupon an overhead presentation was presented as follows.)

CHIEF COUNSEL MONAHAN CUMMINGS: I'm going to do both of the staff updates today. The first one has to do with the other listing processes besides the ones done by this Committee. And we always like to update you and let you know which chemicals have been listed, delisted, or are being considered right now under these other mechanisms.

On the first slide here, you'll see that since our last meeting on May 21st of last year, the office has listed a number of chemicals. Given that I'm not a scientist, I don't like to read off these names, and so that's why we have a slide.

So we had two reproductive -- or two carcinogens that were listed earlier this year. We have this group of chemicals that we call the zines, and some of their metabolites or breakdown products - I'm not sure which way we would want to talk about that - that were also listed.

You'll notice a delayed effective date on that of October the 1st, 2015. I'll explain the reason for that in the litigation update.

Next slide

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CHIEF COUNSEL MONAHAN CUMMINGS: All right. Since our last meeting, we've also delisted a chemical. The chemical name is chlorsulfuron, which used to be listed as a developmental and female reproductive toxicant. It was delisted in June of last year, because of a change in the -- by the authoritative body -- oh, I'm sorry -- and a decision by this committee, I'm sorry. This is yours. You did this.

All right, and then next slide.

--o0o--

CHIEF COUNSEL MONAHAN CUMMINGS: So these are the chemicals being considered currently under our administrative listing processes. I should update you on -- the first one here is nitrate in combination with amines and amides. In some late-breaking news, this set of chemicals is actually being referred to the Carcinogen Identification Committee because of some questions about which actual chemicals were tested and whether or not this category is too broad based on the information that was provided.

So this set of chemicals will actually be heard by the CIC at some -- a future meeting, probably later this year or early next year.

The other chemical that's being currently considered for reproductive toxicity and the developmental endpoint is ethylene glycol. And we published our Notice of Intent to list that chemical in April, and so we have to make a decision before April of next year.

For carcinogens, we have styrene, and then two fairly recent proposals for listing of aloe vera, the whole leaf extract, and Goldenseal root powder which are actually based on the designations by IARC, International Agency for Research on Cancer.

Next slide.

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CHIEF COUNSEL MONAHAN CUMMINGS: We also have currently a proposed safe harbor level for the chemical DINP. And we proposed that safe harbor in January. And it's in the actual regulatory process now. We have to adopt those as regulations. This is a carcinogen. And so the peer review is actually being done by other committee, the CIC. And we expect to adopt a level by the end of the year.

Any questions on the chemicals stuff?

All right. So now I'll put on my attorney hat.

I have a very brief update on litigation. I have to say that we have more cases right now than we've ever had to -- against our office, since I've been here in 13 years.

So just, in no particular order, we have the American Chemistry Council versus OEHHA case. That has to do with a challenge of the listing of the chemical bisphenol A as a developmental toxicant. That case was recently decided by Judge Frawley here in Sacramento in the favor of OEHHA. And the court denied the ACC's request to direct OEHHA not to list BPA. The ACC has filed an appeal of that case. And depending on some procedural things, we may or may not be adding BPA to -- or the endpoint of developmental toxicity to BPA, since you all just listed it.

We'll have to decide -- we'll have to see what the court of appeals says before we do that. Currently, we have an injunction preventing us from doing that.

In the American Chemistry Council versus OEHHA case dealing with, what I just mentioned, the DINP listing, which that was a listing based -- that was done by your sister group the Carcinogen Identification Committee, the ACC challenged that listing. Once again, OEHHA prevailed in the trial court and the ACC filed a notice of appeal on May the 5th.

There's a case called Syngenta versus OEHHA, that is currently in superior court here in Sacramento County. That's a challenge of our no significant risk level or safe harbor level for the chemical chlorothalonil, which is listed as a carcinogen. We're currently -- the status of that case is it's been stayed. We're working on trying to explore a possibility of issuing a Safe Use Determination, and so the case is stayed currently.

Another case filed by Syngenta versus OEHHA has to do with the listing of the triazine chemicals, which I mentioned on the other update. We were challenged on that listing, and we have changed the listing date, pending the hearing in the case, to October the 1st. We have a hearing in September. And depending on the outcome of that hearing, the listing would be effective October 1st, or if the court rules against us, then obviously the chemicals won't be listed.

The last case we have, at least as of this moment, has to do with -- the plaintiff is called the Mateel Environmental Law Foundation -- Environmental Justice Foundation. I'm not recalling at the moment -- versus OEHHA. The challenge is to our current safe harbor level for lead, which was actually adopted in 1989. We have safe harbor levels for lead for both reproductive toxicity and cancer. And this is a challenge to the

reproductive toxicity level.

That case is fairly new. The California Chamber of Commerce and the Farm Bureau just recently intervened in the case, and so we're actually just in motion practice right now at the very beginning of the case. And our next court date is on June the 5th.

Do you have any questions on those?

Now you know why we have two more attorneys working for me.

(Laughter.)

CHIEF COUNSEL MONAHAN CUMMINGS: Thank you.

CHAIRPERSON GOLD: Thank you. I'll turn it over to Lauren Zeise. Sorry.

ACTING DIRECTOR ZEISE: Okay. To summarize the Committee's actions for the day, the Committee had one action, and that was a determination of whether bisphenol A has been clearly shown, through scientifically valid testing, according to generally accepted principles to cause female reproductive toxicity. And the Committee unanimously voted with seven votes, yes. So bisphenol A will be placed on the Proposition 65 list for that endpoint.

Now, I'd like to just say some thank you's. There was a huge amount of evidence for this chemical for that endpoint. And the Committee was clearly well

prepared to evaluate the evidence. And I can't imagine the number of hours you spent working through the literature. So just a very huge thank you to -- for all your work on that, and for taking time out of your really, what we know is, very, very busy schedules to come to our meeting. It's really -- we're really, really grateful.

And I know if George were here today, he'd be very, very pleased with all of the hard work that you've put in.

I'd also like to thank our staff for all the hard work putting together the documentation, for supporting the Committee in their work. Just really a lot of effort goes into preparing for these meetings, and pulling the materials together. So many thanks to staff.

And I'd like to thank the audience that are attending on the web and that came here to participate in our meeting and make presentations. So thank you so much for participating.

With that, I'm going to turn it back over to Ellen -- Dr. Gold.

CHAIRPERSON GOLD: Thank you. I, too, want to thank the staff and the members of the Committee for all their hard work, and the members of the public for their very carefully thought-out statements and adhering to the time frame.

On a personal note, I've worked with George for a number of years and always found him to be very fair and equitable and thoughtful. And I know Lauren will do a great job in his place, but this is a sad day for all of us. And let me just say that I believe we all only found out very recently, and that's why I think emotions are pretty raw. And we wish him and his family all the best.

So thank you all and have a good evening. We're adjourned.

(Thereupon the Developmental and Reproductive Toxicant Identification Committee adjourned at 3:50 p.m.)

C E R T I F I C A T E O F R E P O R T E R

I, JAMES F. PETERS, a Certified Shorthand Reporter of the State of California, do hereby certify:

That I am a disinterested person herein; that the foregoing California Office of Environmental Health Hazard Assessment, Developmental and Reproductive Toxicant Identification Committee was reported in shorthand by me, James F. Peters, a Certified Shorthand Reporter of the State of California, and thereafter transcribed under my direction, by computer-assisted transcription.

I further certify that I am not of counsel or attorney for any of the parties to said meeting nor in any way interested in the outcome of said meeting.

IN WITNESS WHEREOF, I have hereunto set my hand this 20th day of May, 2015.

JAMES F. PETERS, CSR, RPR
Certified Shorthand Reporter
License No. 10063