# Aggregate Exposure Pathway Workshop

**May 9-10, 2016 ▪ U.S. EPA ▪ Research Triangle Park, NC**


## Organizing Committee:

Cecilia Tan (EPA ORD/NERL), Peter Egeghy (EPA ORD/NERL), Stephen Edwards (EPA ORD/NHEERL), Justin Teeguarden (PNNL), Dan Vallero (EPA ORD/NERL), Kristin Isaacs (EPA ORD/NERL), Jon Sobus (EPA ORD/NERL), Paul Price (EPA ORD/NERL), John Wambaugh (EPA ORD/NCCT), Andrew Gillespie (EPA ORD/NERL), Mark Bagley (EPA ORD/NERL), Dan Villeneuve (EPA ORD/NHEERL), Gary Ankley (EPA ORD/NHEERL), David Balshaw (NIEHS Extramural), Michelle Heacock (NIEHS, SRP), Annette Guiseppi-Elie (EPA ORD/NERL)



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# Contents

# Background and Introduction

Recognizing the growing demands for conducting rapid, cost-effective, and reliable exposure assessment on the thousands of chemicals in commerce, a committee convened by the National Research Council (NRC) developed its vision for exposure science in the 21st century. A necessary element of the evolved exposure science paradigm envisioned by the NRC committee is a predictive framework that has the ability to forecast exposures with improved accuracy. To realize this vision, the exposure science community should "adopt a systems-based approach that, to the extent possible, considers exposures from source to dose and dose to source and considers multiple levels of integration."

To enable such a "systems-based approach," the concept of an "Aggregate Exposure Pathway" (AEP) has been proposed to organize data and information emerging from an invigorated and expanding field of exposure science. The AEP framework is a layered structure that describes the elements of an exposure pathway, as well as the relationship between those elements; it offers an intuitive approach to organize exposure information and sets the stage for forecasting exposure at an internal target site (see Figure 1). The AEP is envisioned to be a natural and complementary companion in exposure science to the Adverse Outcome Pathway (AOP) concept used in the toxicological sciences. The direct link between AEPs and AOPs will complete the source to outcome continuum to support more efficient integration of exposure science and toxicity testing information. Successful refinement of the AEP framework, however, requires continuing dialogue and collaboration amongst leaders in the exposure and toxicological sciences.

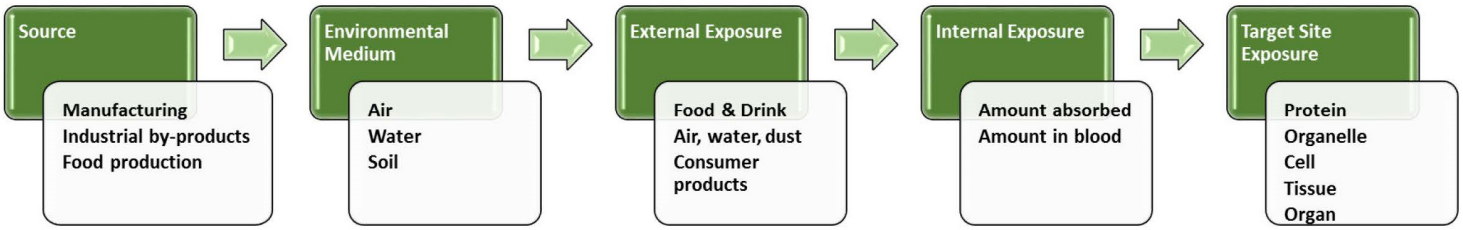



**Figure 1. The AEP: A Structured Representation of Exposure Events**

The U.S. Environmental Protection Agency's (EPA) Office of Research and Development (ORD) hosted the Expert Workshop on Aggregate Exposure Pathway: A Conceptual Framework to Advance Exposure Science Research and Complete the Source-to-Outcome Continuum for Risk Assessment on May 9-11, 2016 at the EPA in Research Triangle Park, NC. This workshop brought together participants from government, academia, and industry to address the following four focused topics:

- Definitions, Applications, and Development of an AEP;
- Infrastructure for Organizing Data and Information in an AEP;
- Integration of AEP and AOP frameworks; and
- Ecological and Human Health AEPs.

Following the two-day workshop, participants were invited to participate in writing teams to develop one or more publications summarizing discussions on these various topics. These publications are expected to promote the development of an AEP framework for organizing information to support public health activities.

# Workshop Summary

The remainder of this report summarizes the presentations and discussions from the workshop. The meeting agenda is included in Appendix A, and a list of registered participants is included in Appendix B.

# May 9, 2016

## Welcome

*Presented by Jennifer Orme-Zavaleta | U.S. EPA ORD/NERL*

On behalf of the EPA's ORD, Dr. Jennifer Orme-Zavaleta welcomed the participants to the workshop and thanked them for attending. She mentioned this was one of many workshops that has taken place in recent years that is addressing advances in exposure science. Dr. Orme-Zavaleta mentioned one of the advantages to the AEP workshop is the combination of risk assessors, exposure scientists, and toxicologists that are thinking through the concept of AEPs. She asked the participants to keep an open-mind in discussing the framework for the AEPs.

## Workshop Overview

*Presented by Monica Linnenbrink | U.S. EPA ORD/IOAA*

Monica Linnenbrink provided an outline for the meeting. She thanked the organizing committee and sponsors from the EPA and NIEHS that helped plan and coordinate the meeting (see Figure 2).

| Sponsors | Group 1<br>Infrastructure | Group 2<br>Applications | Group 3<br>Ecological & Human |
|---|---|---|---|
| • EPA NERL<br>• EPA NHEERL<br>• EPA CSS<br>• NIEHS | • Stephen Edwards<br>• David Balshaw<br>• Kristin Isaacs<br>• Paul Price<br>• Jon Sobus<br>• Yuxia Cui | • Justin Teeguarden<br>• Michelle Heacock<br>• Dan Vallero<br>• Dan Villeneuve<br>• John Wambaugh<br>• Hisham El-Masri | • Peter Egeghy<br>• Gary Ankley<br>• Mark Bagley<br>• Andrew Gillespie<br>• Kellie Fay<br>• Jeremy Leonard |

**Figure 2. EPA and NIEHS Sponsors and Organizing Committee**

## Pathway-Based Predictive Approaches: Linkage between Exposure and Effects

*Presented by William Benson | U.S. EPA ORD/NHEERL*

Dr. William Benson discussed how AOPs can be a starting point for solving many 21st century environmental challenges, such as impacts from climate change, multipollutant exposure, and the thousands of new chemicals in commerce today. There are many scientific challenges in overcoming these complex issues, particularly in generating information to advance the understanding of the complex relationships that exist between human activities and the subsequent impacts to health and the

environment. Dr. Benson discussed that the impetus for this workshop was to acknowledge the interconnectedness of ecological integrity and human health. Another particular scientific challenge is the ability to predict adverse outcomes of societal choices over time and space, which is relevant to our ability to predict the aggregate outcomes and exposures of different stressors. Just as the strong connection between the environment and human health exists, the linkage between exposure and critical effect cannot be understated; we cannot have one without the other.

Dr. Benson discussed "The Great Chemical Unknown," an article published in *Scientific American* in 2010. A small fraction of the compounds around us have been tested for risk, exposure, or hazard. He explained how inefficient the current approach for regulatory toxicology testing remains and stressed that the effectiveness of testing must increase beyond long-term, resource-intensive, whole animal tests. A paradigm shift in regulatory toxicology is slowly occurring, but in order to predict chemical toxicity with limited data, it is important to continue identifying characteristics of normal pathways, develop approaches to predict chemical characteristics that enable perturbations of these pathways, and find a way to translate the mechanistic data generated from these tools into outcomes meaningful to risk assessment.

The AOP framework uses current knowledge on molecular initiation at levels of biological organization relevant to risk assessment. This approach is valuable for a variety of reasons; it establishes common terminology for communication between scientists; encourages a pathway approach that promotes the avoidance of the traditional chemical-by-chemical approach; and is "fit-for-purpose," using weight of evidence-based approaches. Using this pathway-based approach enhances predictive chemical assessments by providing a basis for incorporating alternative or mechanistic data to predict possible adverse effects. It encourages focusing and optimizing of testing needed to assess potential risks by enhancing cross-species extrapolation of chemical effects and by supporting the assessment of effects of chemical mixtures. The AOP concept has been increasingly accepted internationally by the scientific community, including those in the regulatory community, because of how well it informs decision making by encouraging an understanding of biological and physical systems to avoid unintended consequences. Our resources and their effects on natural systems, social systems, and human health and well-being are all interconnected, and we must remain sensitive to these intricacies.

## Q&A/Comments

- **Ron Hines (EPA/ORD/NHEERL):** I just wanted to comment on the concept of the AOP and what it offers. It also offers the ability to look at modifying factors. You can look at the key steps in an AOP and the effect of genetic or epigenetic variation, even for non-chemical stressors. Hopefully that can be integrated into the AEP as well.
    - **William Benson (EPA/ORD/NHEERL):** That's a great point. In making this presentation, I wondered if there would be markers that would be indicated through this AOP process that would lead to better approach, either retrospectively or prospectively, for epidemiology. I think there are a lot of different ways we can think about this. I wanted to mention this potential use of AOPs in retrospective and prospective epidemiology.

## Rethinking Exposure Science

*Presented by Jennifer Orme-Zavaleta | U.S. EPA ORD/NERL*

Dr. Jennifer Orme-Zavaleta began with the importance of pursuing a new exposure science. Without measurements of exposure, toxicology cannot effectively do what it needs to do. Exposure science is important because it guides toxicity testing and risk assessments by connecting the dots between stressors and receptors of disease. More realism is needed in toxicological testing, and exposure science can provide that realism. Pursuing exposure science would result in a better systems approach, one that understands each piece of a system and how they all interact in order to inform better, sustainable solutions. Exposure, both internal and external, is a dynamic system where outcome can affect upstream human and natural factors in a feedback loop. This system needs to be examined at a societal and molecular level, and particularly at the overlap of these two levels.

Dr. Orme-Zavaleta explained that exposure science must capitalize on the major advances that have been made in analytical methods, biological and environmental monitoring, sensor technology, and computational tools in order to move beyond making assumptions about exposure when characterizing risk in order to improve public and environmental health protection. To address the challenges in incorporating these advances, the field of exposure science must move beyond traditional understandings of exposure. This will involve transitioning from our traditional view of exposure, where a certain source leads to a certain disease, to acknowledging and accounting for the complexity of true chemical exposure pathways. Investigation of these systems must change from considering solely point- or route-of-contact as a source of exposure to acknowledging internal doses as sources of exposure; from examining a single chemical or stressor to developing real world, multi-source exposure scenarios; from assessing a single medium discretely to examining multiple sources of exposure simultaneously. Exposure science must also factor in human activity and behavior characteristics and view the entire system in the context of the exposome.

The AEP framework enables us to bridge the full span of information between exposure source and outcome. Dr. Orme-Zavaleta explained the purpose of this workshop was to refine this framework through developing a definition, establishing the infrastructure of AEP, investigating the application of AEP, and integrating ecological and human health AEPs through biological knowledge.

## The Case for Aggregation of Exposure Information

*Presented by Tina Bahadori | U.S. EPA ORD/CSS*

Dr. Tina Bahadori discussed how the AEP fits into the near-term strategic plan for ORD. She began by explaining that the field of exposure science has advocated for better integration in risk assessment but has been repeatedly marginalized. This marginalization is a result of investigating hazard and exposure separately rather than acknowledging the interaction between the two.

There is an increased flow of exposure information in recent years, including novel data and publications regarding monitoring, modeling, non-targeted analyses, product information, informatics approaches, and better reporting. Without an effective way to organize the data, it is difficult to fully understand the impact of exposures; address research gaps and apply resources; and mitigate detrimental exposures or enhance beneficial ones.

With the introduction of AOPs, biology has become relevant again. Exposure to chemicals must be understood in the context of reality, not just those in a laboratory setting. Without the information we

need to fully characterize exposure, biology can lay the groundwork for predictive approaches by presenting putative and plausible models to evaluate the impact of exposures.

Dr. Bahadori discussed that the Teeguarden et al. (2016) paper[1] shifted the conversation from exposures we know about to incidental, chronic, low-dose exposures and their importance over our lifespans. If nothing else, these exposures contribute to the chemistry and biology of human health, and we must understand each of these to fully understand an exposure's impact. Of particular note are the synergistic but not simultaneous exposures that occur over space and time. Cumulative risk assessment attempts to understand this relationship of mixtures, but we must move beyond mixtures of chemicals to investigating stressors at low doses in different life stages and how these contribute to the long-term effects of a system. She discussed the disruptive potential of mixtures, arguing that the AOP has begun to elucidate the nuances in chemical effects. For example, a chemical may not be a carcinogen in and of itself, but it may contribute to the disruptive pathway that eventually leads to carcinogenic effects.

Without fully understanding the characteristics of stressors, it would be difficult to generalize the characteristics and roles of exposures that lead to carcinogenicity. In order to be more preemptive of harm, we must better understand relevant exposures by better understanding the biology and chemistry of exposures in health. Previously, our regulatory approach has been based on calculating acceptable risks from risk assessments informed by hazard evaluations that are reliant on traditional toxicology. The public health approach contrasts with many aspects of the regulatory approach, considering the agent, host, and environmental framework together. Public health has focused on prevention and health promotion, particularly in susceptible and vulnerable populations, emphasizing eliminating risk and improving overall health. By shifting our view more towards the public health approach, we can place more emphasis on what we understand and what we can prevent. The AEP framework can take us in that direction, as it is a tool to organize the available information in order to guide us to where we must take action.

Transformation in the field of exposure science necessitates building upon existing knowledge. Linking the AOP with the AEP will help us build upon our understanding of current exposure data. Exploiting complex systems modeling to advance mechanistic understanding will help integrate our understanding of exposure and dose effects across multiple levels of biological organization, helping us to predict early "tipping points" that matter for human and ecological health. Considering exposures that matter, such as those that affect vulnerable and susceptible populations and life stages, including accounting for early life exposures with long-term health implications, should be of particular importance. Furthermore, investigating chemicals will only elucidate one piece of the greater exposure system puzzle. We must re-think what cumulative exposure and cumulative risk assessment mean, and greater knowledge of biological systems will help us better understand the full picture. Through the integration of this knowledge, exposure science will become a more effective tool for protecting human health.

[1] Teeguarden et al. (2016). Completing the link between exposure science and toxicology for improved environmental health decision making: The aggregate exposure pathway. Environ Sci Technol 50(9):4579-4586**:** http://pubs.acs.org/doi/abs/10.1021/acs.est.5b05311**.**

## Q&A/Comments

- **Glenn Suter (EPA/ORD/NCEA):** You said that the Agency regulatory approach is based on risk assessment which is informed by hazard, but not exposure. However, we regulate exposure – we regulate concentrations in various media and uses, so how can this be true?
    - **Tina Bahadori (EPA/ORD/IOAA/CSS):** We have not used exposure to guide the need for regulation. We have done hazard identification first, and if the hazard has raised a flag, then we use exposure to control those hazards. This is different than the interaction between exposure and hazard being used to drive the regulatory decision making. We have had very little exposure information.
- **Glenn Suter (EPA/ORD/NCEA):** You say that "chemicals cannot interact simultaneously?"
    - **Tina Bahadori (EPA/ORD/IOAA/CSS):** Not cannot, but "do not necessarily" interact simultaneously.
- **Weihsueh Chiu (Texas A&M University):** One of the things I think you were saying with the cancer example is that, even with AOPs, there is an overemphasis on the molecular initiating events and perhaps even that is "looking under the lamppost." But in fact, all of the nodes in the pathway are important and potentially may interact with chemical or non-chemical stressors. Maybe having a more equal footing across the whole network of nodes would actually be better – is that what you're saying?
    - **Tina Bahadori (EPA/ORD/IOAA/CSS):** I think that's what that paper made me think about. We need to quantitatively develop AOPs so they can be used. We have to look a little bit under the lamppost so we can quantitate and describe them. But in order to link that to the reality of the biological outcome, totality does matter. We need to look at the balance of the role of the other nodes of the network as well. At least in the case of cancer, that is the case.
    - **Weihsueh Chiu (Texas A&M University):** I think that also gets at Ron Hines' previous comment that they reveal where other parts of the pathway might be modified, but what is the modifier and what is the initiator? It is dependent on your point of view.
    - **Tina Bahadori (EPA/ORD/IOAA/CSS):** Thinking is going to have to be iterative. As we get more data and as models are more developed, we can understand that network better, too, and I think that is the goal ultimately.
- **Christine Hendren (CEINT):** Do you see this whole concept rolling out as an education piece for people who are toxicology-focused, hazard-focused, and exposure focused? Or do you think that there needs to be a new type of professional whose career would live at this intersection?
    - **Tina Bahadori (EPA/ORD/IOAA/CSS):** I would argue with you that if you look at almost any young scientist these days, they already come at it from an integrated perspective and looking at exposures that are operating in the effect space. The people struggling are my generation, and we control the purse strings at the moment. So, my generation needs to think about how these new models exist. In fact, there's a paper in Environmental Health Perspectives (EHP) that just came out in advanced publication co-authored by Linda Birnbaum, Tom Burke, and Jim Jones that looked at the real divide that exists in chemical space between environmental epidemiology and risk assessment. One of the biggest contributors to that divide is the funding mechanisms. The funding mechanisms that have not allowed the transdisciplinary science that we need and that we all talk about to actually develop. I agree with you, there is a mind-shift that needs to occur. As we see the promise of these integrative approaches, I think even the older generations will be able to see the light.

- **Christine Hendren (CEINT):** I wonder if you could also work with professional organizations that work with funding agencies and also academic administrators to try to change incentives to make it beneficial rather than a taxation to work cross-disciplinary.
    - **Tina Bahadori (EPA/ORD/IOAA/CSS):** Thank you, that is a good idea.
    - **Justin Teeguarden (PNNL):** I think Tina Bahadori mostly made my point and addressed my question, but maybe I am a little less optimistic than you are. You are viewing this from the perspective of within the EPA, where there are a lot of young folks doing some dynamite stuff. Part of your mission is to integrate those data streams and those sciences. Outside of EPA, however, where the funding agencies are not driving for interdisciplinary research as much as I think they could, you are faced with separation of toxicology, epidemiology, and exposure. The overlap is not happening as much as you would hope. You walk down the way of posters, many of them funded by federal agencies at the Society of Toxicology (SOT), and it's still micro- and millimolar doses. You ask people about the exposure context, how it affects your mechanism or exposure biology, and they are clueless. Penetration may be good at the EPA, but it is not considered in academic community. It is not driving their interdisciplinary research and training of younger people.
    - **Tina Bahadori (EPA/ORD/IOAA/CSS):** Training is a big part of it, I agree. If we end up embracing the AEP, we're going to have a few more workshops to talk about how we are going to operationalize it. You have very valid points.
- **Nicholas Anastas (EPA/NRMRL):** How does this AOP/AEP concept fit with the existing Risk Assessment Guidance at Superfund Sites (RAGS) that considers both hazards and exposure?
    - **Tina Bahadori (EPA/ORD/IOAA/CSS**): There is a lot of learning to be had from what has happened under the Resource Conservation and Recovery Act (RCRA) and Superfund because their exposures have mattered. But even there, the issue has been that we have done the exposures fairly separately from assessment of the risk. Going back to what Glenn Suter was alluding to, I can see that we continue to disagree on this, there is a lot of learning there. To set up a superfund cleanup strategy, we begin by quantifying the chemicals we know how to measure, we quantify their levels and their hazard or toxicity properties, and that defines our risk assessment strategy. We have found that had we taken a non-targeted analysis approach and looked at the chemical soup at a typical Superfund site (if such a thing exists) our outcome strategy may have been very different. Understanding the chemical and the chemicals we know how to characterize has overshadowed the importance of understanding the relevant exposures. That being said, at least Superfund looked at exposures. There are a lot of learnings to be had there, but we have to build on it and begin to do a better job of mapping relevant exposure to relevant stressors, their interactions, and how we need to quantify them.

## Introducing Aggregate Exposure Pathway (AEP)

*Presented by Cecilia Tan | U.S. EPA ORD/NERL*

Dr. Cecilia Tan began by describing the Teeguarden et al. (2016) paper on the AEP[1]. This workshop is the first regarding the AEP, and the goal is to produce additional manuscripts and more complete guidance to address the use of exposure tools for developing AEPs. Although the exposure pathway concept is not new, it can be incorrectly used as a synonym for exposure route or exposure scenario. The AEP concept is similar to that of the Conceptual Site Model, which illustrates the links between environmental contaminants to human and/or ecological receptors.

To first move forward, Dr. Tan discussed the necessity of refining the "exposure pathway" concept through the use of a generic framework. This new framework must include a concrete structure and common nomenclature and enable better integration with hazard information by taking into account sources to internal targets and informing the exposome with both chemical and non-chemical stressors. The exposome comprises every exposure an individual experiences from development to death. While the AOP can provide a structured representation of biological events, the AEP can provide a structured representation of exposure events. Both consist of two basic elements: key events (KEs) and key event relationships (KERs). While the AOP connects a molecular initiating event and an adverse outcome without explicitly including chemicals, the AEP connects the source and target site exposure without explicitly including toxicity. Using the AEP and AOP together allows for deduction of responses to chemicals and other exposures.

Dr. Tan then provided an example of a stressor-centric AEP, focusing on the transformation of chemicals leading to different AEP, which when placed together formed a network. The granularity of the KEs in the AEP depend completely on the application, demonstrating how AEPs can be fit-for-purpose. She also demonstrated how adding an internal exposure can add complexity to the network. Additionally, Dr. Tan raised various questions that she hoped would be answered throughout the workshop:

- How should exogenous and endogenous sources in AEPs be considered?
- Should exposure factors, physiological characteristics, and pharmacokinetic characteristics be considered KEs or KERs?
- Though AEPs will be stressor centric, how can they be modular?
- How can individual AEPs be built, and would it be the equivalent of an AOP network?
- What would an AEP network look like, and what applications would an AEP network have?

AEPs are planned to be living documents, and these thoughts and questions are hoped to guide AEP development at the workshop. Dr. Tan explained that there is a need for an infrastructure for AEPs that allows users to associate features with exposure, understand real-world mixtures, inform toxicity testing, support source mitigation, and identify green alternatives. In addition, there is a need for a place to store and organize all of these data, or a place where these features can be mapped during non-targeted analyses.

## Q&A/Comments

- **Harvey Clewell (ScitoVation):** I can see a lot of potential for this, particularly if you have feedback so that you can look at if you should put certain chemicals together in the exposure assessment. As a modeler, this won't be useful unless it is actually supporting the development of models that will then incorporate all of these things in a way that people can implement and apply in order to get quantitative results.
- **Ron Hines (EPA/ORD/NHEERL):** AEP's are stressor-centric – does that mean chemically agnostic as well?
    - **Cecilia Tan (EPA/ORD/NERL):** Justin is shaking his head no. We had that discussion three different times in three different workgroups, and that is the question we'll have this afternoon: can it be modular? Can it be reused?
    - **Justin Teeguarden (PNNL):** There is a partial answer there. For the most part, we see them as being chemical centric, because you have to talk about the pathway of a stressor to some place. You're not describing the basic biology as you are in an AOP. There are cases, where there are boxes or key elements that can be reused. So for example, if you had a box that described the metabolic capacity of a P450 system as a key transformation

before you deliver a reactive metabolite, it may be for chemical 1, but you may also be able to use that for chemical 2. Or, if you had a box in the environment that dealt with the oxidative potential of a particular soil type at a certain site, that also could be used for a class of chemicals that are also oxidizing – polycyclic aromatic hydrocarbons (PAHs) or something – to a chemical of concern at the end. So we're not fully sure if it is one or the other. In my opinion, I do not think it is agnostic.
    - **Cecilia Tan (EPA/ORD/NERL):** Another example would be air – the whole box can be chemicals in the air, and you can separate them based on a single chemical.
- **Stan Barone (EPA/OPPT):** I want to see the implementation. One of the challenges that we talked about in Washington was that we're dealing with multiple chemicals, multiple classes of chemicals, and multiple media. I want to know how we are going to deal with dust; fish; classes of chemicals, aggregate and cumulative? How do we have this work together for a better exposure assessment?
- **Ron Hines (EPA/ORD/NHEERL):** My concern is that if you make it chemical specific, you drown in the same problem of looking under the lamppost and having 150,000 different aggregate exposure pathways to deal with. Unless we have a way to use modular systems, and try to aggregate the chemical classes (e.g., common PBPK features), we're going to drown in a lot of information we just can't handle. I would urge us to take a broader approach and look at how to group things.
- **Elaine Cohen Hubal (EPA/ORD):** How do we avoid getting lost in the weeds? I see the KEs, which I see as key determinants, being a box that would hold some sort of generalizable data. Any particular chemical can be applied using the arrows and the generalizable number from the box to lead to a slightly different answer.
- **Olivier Joliet (University of Michigan):** Optimistically, there is a tradition if you look at environmental exposure to have this box-feature where multimedia models are run for 3,000 chemicals, but structure of prediction mostly the same between chemicals. I think the PBPK approach, which has been more traditional but also more chemical specific, can also be formulated. Mathematically, all of that can be formulated into a framework, which should be relatively easily adaptable. I'm relatively optimistic that we can find this sort of structure, which I think exists more in the environmental world than internally.
- **Rory Conolly (EPA/ORD/NHEERL/ISTD):** In the AOP world, much of work has been done at the qualitative level. In regards to AEPs, what do you think of them in terms of quantitative vs qualitative tools?
    - **Cecilia Tan (EPA/ORD/NERL):** There are various stages of development for AEPs similar to AOPs. For a lot of chemicals out there, there is no toxicity data or no information whatsoever, but you know that it exists around you or in the body. I would imagine these situations would be qualitative, putative AEPs.

## Infrastructure to Support AEP Development Lessons from the AOP Knowledge Base

*Presented by Stephen Edwards | U.S. EPA ORD/NHEERL*

Dr. Stephen Edwards provided background on AOP development, which began in 2012 by the Organization for Economic Cooperation and Development (OECD) to serve as a knowledge base. The success of the AOP was largely because there was an international need to both build a driver and use a knowledge base. Guidance and training are available for users of the AOP. Currently, there are 20 AOPs under review, with about 100 more under development. Dr. Edwards then discussed the components of

the AOP Knowledge Base (AOP-KB): the AOP Wiki, AOP Xplorer, Effectopedia, Intermediate Effects DB, and some Third Party applications and plug-ins, all of which can be found in the AOP-KB Hub. Each of these modules allows data exchange among and between the others.

This AOP-KB supports all stages of AOP development: putative, using AOP explorer to aggregate information; formal, using the AOP wiki to focus on evaluation; and quantitative, using Effectopdia to create very explicit and detailed AOPs with the intention of use for computational purposes. All of these stages preceded the knowledge base, but have been modified for use while still keeping true to the original purpose of the individual tools. AOPs that share KEs naturally form AOP networks, which are intended to build up over time and are stored in the AOP-wiki. The communication among these diverse systems allows for this network to build.

Dr. Edwards continued his presentation explaining that all of the descriptors of the AOP can be seen as analogies for the AEP: the creation of sub-networks that exchange information across pathways to examine aggregate exposure and cumulative exposures. Many AEPs will lead to a few toxicities. The measurement of cumulative risks, from chemical and/or non-chemical stressors does raise questions regarding how to deal with target site exposures in the same location. In addition, AOPs are typically chemical agnostic, but the AEP will require the need to think about how these chemicals or stressors affect metabolism. The connection of AOPs and AEPs is also still in question. By creating ontologies that describe KEs in existing AOPs, as well as the other lessons learned from creating the AOP-KB (such as developing a driver, the actual design, and the technical challenges with building federated systems), how do we create an effective AEP infrastructure? In particular, is there a need in the scientific community that a common infrastructure can address? Which existing resources can be leveraged? What should be stored in the module, and can we create a module that can handle that volume of data and provide support for analytical workflows and/or needed queries? Information must be standardized and exchange protocols must be established to develop an effective AEP Framework.

## Q&A/Comments

- **Weihsueh Chiu (Texas A&M University):** AEPs seem to have an advantage over AOPs. All the existing infrastructure is based on existing quantitative models, essentially. As a modeler, I always think in terms of states and the processes that define the relationship between states. The idea of a KE, which has the connotation of something that is discreet, may not be the right nomenclature for exposure because exposure is really about a continuum of different state variables. In terms of AOPs, I think we should be thinking about states instead of KEs. Specifically, we should try to define the boxes as the state of the biological system at various levels of organization and then identify the processes that relate changes in states as opposed to thinking about it discreetly.
    - **Stephen Edwards (EPA/ORD/NHEERL):** AEPs and AOPs share more than you would think. In the exposure world, you are already dealing with this area. It should come more naturally to exposure scientists. Part of the definition of a KE is that it represents a change in biological state. The meanings aren't that different. The dynamics in AEP and AOP fall in the arrows. It's not that you're dealing with discreet quanta; rather, it is something you can measure, or a way of interrogating the system.
    - **Weihsueh Chiu (Texas A&M University):** To use cancer as an example, the state of system is that there are always some initiated cells. It is not that something comes in and then there is initiation. It is more of a change in overall rate of initiation, or fraction of those cells initiated at a certain time and is really more of a continuum. This also brings

into question the variabilities and susceptibilities, which differ between people, and also their baseline states, which varies through life stages.

- **Stephen Edwards (EPA/ORD/NHEERL):** I can't get into modifying factors on AEPs in this talk, but we should talk later.

## Potential Applications of the AEP Framework

*Presented by Justin Teeguarden | Pacific Northwest National Laboratory*

Dr. Justin Teeguarden began by explaining that this workshop aims to improve public health through better integration of exposure science, toxicology, and epidemiology. With an organizing framework that allows archiving and efficient use of data for prediction and decision making, public health improvements can occur more quickly and completely, leading to a more immediate impact. Access to more data will necessitate a way to deal with this increased amount of data. Depending on the way this organization of exposure data is built, it can have a better tie to both the toxicology and epidemiology communities.

Before discussing this, however, Dr. Teeguarden stressed that we must discuss AEP applications. Without applications with meaningful uses for this type of framework, there is no reason to develop the infrastructure. Ontology and terminology must be responsive to relevant applications, and applications must be clear so as to avoid developing a flawed design. We want to emerge from this workshop with more market drivers for development. Without the application discussion, however, we cannot possibly predict pitfalls and plan how to avoid or overcome these pitfalls. Dr. Teeguarden discussed some generic applications of the AEP, most importantly of which is that the AEP will drive improved integration of exposure science with other public health disciplines. It will also be useful for data acquisition, organization, and mining; the integration of the AEP and AOP will be helpful for public health decision making; it will help scientists to aggregate exposure pathways by source, chemical class, route, pathway, or molecular initiating event; it will be useful in cumulative risk assessment; and it will encourage the accumulation of the right type of data to both understand and execute exposure modeling well.

Dr. Teeguarden provided some examples of how the AEP could potentially be used, including measuring total estrogenicity from multiple sources in order to weight and mitigate the exposure pathways of highest impact, or the identification of source attribution and exposure data for future scientific testing that could lead to better risk assessment or management decisions. He also discussed that this workshop is an excellent opportunity to reshape how exposure data is acquired, assembled, and consumed in public health.

## Integration of Human and Ecological AEPs

*Peter Egeghy | U.S. EPA ORD/NERL*

Human health and ecological risk assessment methodologies were originally developed independently; however, the idea of integrating the two quickly followed. With the integration of the risk assessment methodologies, there was an expectation for better decisions when risk to both human health and nonhuman organisms are considered. The need for an integration between ecological and human health risk assessment emerges from the interdependence between ecological and human health risk, the need to avoid duplication for common processes, and the idea that nonhuman organisms can be more highly exposed, more sensitive, and more easily sampled. Overall, coherent assessments aid better decision-making processes.

Human health and ecological risk approaches are different in many ways. Some of these differences include: 1) ecological conceptual models including routes of exposure not relevant to humans,

2) ecological considerations going beyond individual organisms to population, community, or ecosystem levels of organization, 3) ecological conceptual models considering indirect effects such as loss of food resources or habitat structure, 4) ecological assessments tending to be more site-specific with fewer default parameters, and 5) ecological assessments reflecting differing temporal scales.

Integrating human and ecological systems means human and ecosystems are viewed as a single, larger, integrated system, with a focus that extends beyond the one-direction effects of ecosystems on humans. Integration would address human effects on ecosystems and even ecosystem disturbances with no impact on any human receptors with incorporation of possible indirect effects. Ultimately, integration would allow for assessment of a broad range of ecosystem stressors beyond chemicals that perturb the natural functioning of ecosystems.

There have been previous frameworks for integration such as the EPA's "Framework for Cumulative Risk Assessment" released in 2003, but integration still remains a goal today. Currently, there is a disconnected application of exposure science to human health and ecosystem health due to insufficient recognition of the connection between the two. AEPs develop a generic framework to provide the following:

- Common features that are familiar and easily adaptable,
- Consistency across different models, allowing for their integration, and
- Rigorously defined components to harmonize model evaluation.

Dr. Egeghy presented pre-workshop discussions and questions to the workshop attendees that must be considered when discussing the AEP framework:

- **Terminology**: Does the terminology need to be consistent with AOPs? Is the terminology consistent with respect to flows and masses?
- **Non-Chemical Stressors**: Synergistic interactions with chemical stressors may exist at the site of action. Some non-chemical stressors may modulate the exposure pathways, some may modulate toxicity pathways, and some may simultaneously modulate both exposure and toxicity.
- **Beneficial outcome pathways**: An example of a beneficial outcome pathway is fluoride in water which exemplifies the need to balance beneficial versus harmful effects.
- **Linear framework**: Ecological interactions may be cyclical, and ecosystems are not linear.

To explore integration between human and ecosystem health, Dr. Egeghy suggested case studies may be useful to explore the framework more in depth. The proposed case studies included the following:

- Linear Akylbenzene Sulfonate (LAS) or methylparaben in shampoo
- Perchlorate contamination in ground/drinking water at Mount Rushmore
- Thiazopyr (herbicide)
- Polychlorinated biphenyls (PCBs) in Bellingham Bay from a defunct paper mill
- Diclofenac for treating cattle

## Workshop Participant Discussion

*Led by Cecilia Tan | U.S. EPA ORD/NERL*

- **Harvey Clewell (ScitoVation):** We must think about how to integrate this in a mathematical or quantitative way. There is one big difference between AEPs and AOPs: AOPs rely on cellular information, but that is relatively new knowledge from new technology. Mapping environmental models dates back to the 1970s and 1980s and continues into today. We are actually ready to

implement exposure information in quantitative models. So we want to be sure that the qualitative world should be amenable to the quantitative world to make sure they use it. So if we are talking about the source to receptor part, obviously it needs to be incorporated into the Stochastic Human Exposure and Dose Simulation (SHEDS). That is a connection that needs to be kept in mind all the time. There are tools that have been around. For example, ERDEM (Exposure Related Dose-Estimating Model) was developed 30 years ago, and now there is population life course exposures to health effect model. Old models are now coming back. We should be sure to think about the tools while we're also thinking about names.

- **Stan Barone (EPA/OPPT):** Both on the existing chemical and new chemical side, one of the challenges we have in exposures is that because we generally have lack of data, we're using generic scenarios and generic models to do our predictions. It's not just about going out and measuring a lot of stuff, which is the old way of doing things. Risk assessors are going to want to know how all of that information generalizes. How do we genericize things so we can use this information in more facile ways?
    - **Justin Teeguarden (PNNL):** I don't have a complete answer to that, but it seems related to what Harvey Clewell said. If in the past, people measured in a box (air/soil), they were content. If we push this new idea of the AEP, people will be forced to think about boxes up and down stream, which should put them in the position to think about states and change, which should make them think about measuring the things in those boxes in a quantitative fashion across some time so that information becomes available for modeling and genericizing.
    - **Stan Barone (EPA/OPPT):** If you move to risk assessment side and think about how we incorporate this into the risk assessment and think about the overlapping pathways and effects as well as exposures and releases, you realize that these are not mutually exclusive pathways. We can't just measure each box and forget about the total exposure, the exposome.
- **John Wambaugh (EPA):** One contrast with AOP, which as long as it is taken into account is just a feature and not a bug. When I think of AOPs, I think of something like thyroid hormone access functions. I presume my thyroid hormone access functions pretty much like my grandfather's did. But with AEPs, there is an element of timeliness. You can have perfect knowledge of the composition of a can of new coke, and that's almost irrelevant to those who are drinking it later. On the other hand, the PCBs produced at that same time are still relevant today. So with each of these KEs, we need to keep in mind "what's the expiration date on that information? What's the relevance to today?"
    - **Cecilia Tan (EPA/ORD/NERL):** We did talk about that in one of the workgroups – production volume changes from year to year. The time component must be addressed, particularly how to capture it.
- **Rick Lippin (Preventative Medicine Advocate):** I'm a physician. I did hear some references to estrogenic effects this morning. I want to put a plug in for the voluntary and involuntary exposure all of us have from ingesting pharmaceuticals. I'll leave it at that for inclusion in future discussions.
    - **Justin Teeguarden (PNNL):** That is a great example of working backwards from exposure driven by an AEP. If you've got an adverse outcome that you know is related to pharmaceutical exposure, but there are other dietary contributors to that same particular pathway that we need to know about, there is value in assembling the exposure information to understand how all these things are working together. Maybe even change dosing for medications, as an extreme example.
    - **Tina Bahadori (EPA/ORD/IOAA/CSS):** That's valid if you are taking the medication intentionally. If the exposures are through your drinking water, they are not intentional.

- **Carolyn Mattingly (N.C. State University):** I just want to put in a plug for the Comparative Toxicogenomics Database (CTD). It was previously mentioned that there wasn't a resource out there to bring together exposure information, but we're starting to build the foundation by specifically curating exposure data and trying to pull it in to other molecular data that is maybe more relevant to the AOP side of things. I would encourage people to take a look at the database now. The query capabilities are not quite there quite yet, but snapshots of data are available. I'm hoping we can get some feedback from people here because we can certainly adjust what we're capturing and how we're presenting it to help it feed into this effort.
    - **Tina Bahadori (EPA/ORD/IOAA/CSS):** There is a lot of effort that has gone on to curate this data into one place.
- **Rick Becker (ACC):** One lesson I learned from working on AOPs was that it is really important to think about applications during development. We heard some of that earlier with some of the examples. Even in the time we spend thinking about definitions, we should keep in mind some applications in order to hone what's actually practical and what can be incorporated in the short-term. In the long-term, you can build a modular-basis and build on to that. Also, in terms of application, maybe think about things in a tiered, iterative manner. We do this naturally when we apply models or approaches – we look at what is found in the screening level approach, and then we move forward if we can't get enough information from that level, which gets into problem formulation. We need to think about both problem formulation and application as we design this effort. Having some of those example up front will help us truth-test some of this as we go forward in the discussions in next few days.
- **David Balshaw (NIEHS):** Thinking at a higher level about the AEP and AOP framework, and the integration of the two, a word that's been used a few times across the earlier presentations was "agnostic/discovery/untargeted." What got me thinking about this was Justin Teeguarden's slide with cumulative load of estrogens. We can use that as a framework for AEP/AOP integration, but one of my concerns with doing that is how to deal with off-target effects. Off target effects of pharmaceuticals or environmental factors actually dominate the adverse biological response to those things. If you focus it on what you know (the estrogenic load) you have a very high probability of missing what's actually important. Is there a way to use AEP/AOP framework in a discovery mode where you are not using pre-information to drive development of pathway?
    - **Justin Teeguarden (PNNL):** Absolutely yes. If we start building now, the system grows, the amount of exposure information grows, and the AOP network will be impacted by AEP targeted and non-targeted effects. Over time, as the exposure biology comes to play and that gets put into these databases, then we'll be able to see dominant roles of those non-targeted effects emerging from pathway.
- **Michelle Embry (ILSI HESI):** There was a cumulative assessment done a few years ago on perchlorate in drinking water. It was found that T4 deficiency in pregnant women was a big problem, and there was a big analysis, subsequently. It's very interesting report that integrates public health, and it gets at what the U.S. Food and Drug Administration and EPA regulate. I also wanted t to get people thinking about the field of exposure science and the current push towards collecting more data. One question I've had while listening to the AEPs is the role of modeling and modeling data. Where does modeled data fit in to this process? How does this fit into the development of AEPs? Maybe gets at the previous point of a tiered approach. I think the focus is always on data, but in exposure modelling it is a big piece.
    - **Stephen Edwards (EPA/ORD/NHEERL):** In one of planning work groups, we talked about that very question. Whereas on the AOP side, the knowledge base tends to focus on measured data, and the modeling builds on this. The consensus of the smaller group is

that this probably will not be the case for AEP and will likely be where the two diverge. Comes with lots of challenges that hopefully we'll address over the next few days.
    - **Justin Teeguarden (PNNL):** I would say that the AEP is not to be constraining. Toxicology and exposure science are not the only fields that use modeling and simulation to create knowledge (whether you call it data or not). I would expect that those activities are supported by the kind of data we collect. I can imagine a scenario where we can create that knowledge if we have enough to build upon. However, there are a lot of people who don't think that is real data. They assume that it cannot be trusted if produced in any way but the traditional approach. This will be part of the AEP development.
    - **Tina Bahadori (EPA/ORD/IOAA/CSS):** The purpose of gathering should not be to consider thoughts of our least common denominator. We should strive to do better than that.
- **Rory Conolly (EPA/ORD/NHEERL/ISTD):** I'm trying to understand what a computer model of an AEP might look like. I work on models for AOP, which describe biological systems and very well might extend over many levels of a biological organization. Conceptually, it's like a box that describes biological entities with quantitative relationships between the boxes, producing a flow. The analogous thing in the environment for how a chemical would move through the environment to a reach an organism might be a geospatial model. Is this what we mean for AEPs? Is this what you expect? Do we mean to identify the key components of the environment, as well as how and why chemicals move the way they do to affect organisms?
    - **Justin Teeguarden (PNNL):** Yes, that's something we talked about. We can imagine that instead of driving research to study chemical concentrations in soil alone, we should really study the capacity of a soil compartment or an air compartment to make a oxidative transformation so that then, in turn, becomes the focus of research and can become applied to similar chemicals. Then, of course, there are space and time and behavior to take into account, so yes, I think all of those are appropriate targets of information.
    - **Tina Bahadori (EPA/ORD/IOAA/CSS):** Far-field modelers have addressed this, but near-field have not yet addressed these issues.
    - **Rory Conolly (EPA/ORD/NHEERL):** The point of developing AEPs like that is that they could become chemical agnostic.
    - **Justin Teeguarden (PNNL):** Yes, they could.

## Parallel Workgroup Discussion on "Definition and Development of AEP"

Parallel workgroups addressed the following three charge questions. High-level summaries from each of the three workgroups are presented in this section.

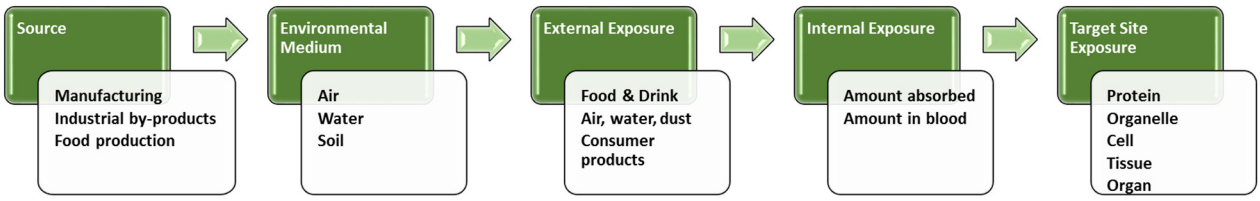
Charge Question 1. The Aggregate Exposure Pathway (AEP) concept is proposed to be the assemblage of existing knowledge concerning biologically, chemically, and physically plausible, empirically supported links from introduction of a stressor into the environment to its concentration at the site of action. An AEP may represent multiple sources and transfer through interconnected pathways that all merge on a single internal site of action, or a single source through one or more pathways merging on a single internal site of action, or any combination of these.

- How would you define an AEP?
- Can a single AEP represent multiple sources of a single chemical to multiple biological targets?
- How would you define multiple AEPs that share the same KE, e.g., a non-specific metabolite generated from exposures to multiple chemicals?

- What existing concepts such as an "exposure pathway" should be considered when defining an AEP?
- Should AEPs for non-chemical stressors or "endogenous exposures" present different considerations vs. chemical stressors?

Charge Question 2. An AEP has elements that are similar to those of Adverse Outcome Pathways (AOPs) in that the basic building blocks of an AEP retain the naming conventions used for AOPs: KEs describe the measurable, obligate steps through the AEP (green boxes in the example figure below); and KERs describe the linkages between KEs (green arrows in the example figure below).

- Should all AEPs have the same types of KEs (i.e., source, environmental medium, external exposure, internal exposure, and target site exposure)?
- How would you define KEs (e.g., location and concentration of a stressor)?
- How would you define KERs (e.g., linking one KE to the next, or representing some transformation such as fate, transfer, absorption, distribution, metabolism, excretion)?




Charge Question 3. Five core principles were proposed for the development of AOPs: AOPs are (1) not chemical specific; (2) modular; (3) a pragmatic unit of development and evaluation; (4) AOP networks are the functional units of prediction; and (5) constantly evolving "living" documents. Where the AOP framework was fit to a single purpose of understanding the pathway from a molecular initiating event to an adverse outcome, the AEP framework may serve several purposes, such as understanding source contributions, or supplying target site exposure information for linking to AOPs. Thus, the core principles for the AOP development may need to be modified and extended.

- What are the core principles you would propose for the AEP development?
- Are there principles that are too constraining? Do any rule out key applications?
- Do any key principles pose undue burden to an application community (e.g., ecological health or epidemiology)?
- What would be the AEP equivalent of an AOP network?

### Workgroup 1 Discussion Summary

Dr. Stephen Edwards led the group discussion for this workgroup and asked for an emphasis in the definitions to help make the AEP useful as well as address the infrastructure information that would be discussed more on the second day. Overall, the group had an in depth discussion pertaining to the components of the AEP to try to determine an applicable definition.

Many challenges arose when trying to determine an overall definition for the AEP framework in workgroup one. Some of these challenges included:

- Whether the AEP should go from source to target tissue
- What the best starting point for AEPs should be
- Whether it is necessary for the AOPs and AEPs to always be linked?

- How the different disciplines will help contribute to the AEP framework
- How the AEP framework will be helpful in the short-term to all disciplines in the exposure process
- Whether AEP will be a research tool or a regulatory tool: Different audiences may have different levels of use and capturing this in the definition is important
- Where to get started on aggregate exposures from external exposures: If the AEP is a repository source for the data measurements, how does that go upstream to understand the external sources
- Overall purpose for the framework which will shape the definition
- Terminology/nomenclature; what standard language will be used so that all different communities will understand:
    - There has been a lot of work done in the past 20 years that would be helpful in the framework (especially in the ecological field), but language is not the same, so communication is lacking between different disciplines
    - Should this framework be called pathways or networks?
- Difference between a state and event
- Measurability of the KEs/thresholds:
    - It was suggested to have this preserved in the AEP and that this would make the framework more useful
- Should the AEP be chemical agnostic or chemical specific?
- Should the terminology for AEP match the AOP?
- How will the AEP help with nontargeted measurements?
- Temporal-spatial dynamics:
    - How would one capture all of this information in the framework?
- Is there a reason to keep these as linear pathways?
    - Some members of the group would prefer that these are linear pathways for ease; this makes it easier to enter and record the information

The overall definitions the group agreed on are as follows and were presented during the Workgroup 1 report out to the larger workshop audience:

- **Exposure Pathway**: 1 source → 1 target site exposure (see Figure 3)
- **Aggregate Exposure Pathways**: multi-source, single chemical → 1 target site exposure (see Figure 3)
- **Cumulative Exposure Pathways**: multi-source, multi-chemical → 1 target site exposure (see Figure 4)
- **Cumulative Exposure Network**: multi-source, multi-stressor → multi-target site exposure (see Figure 5)

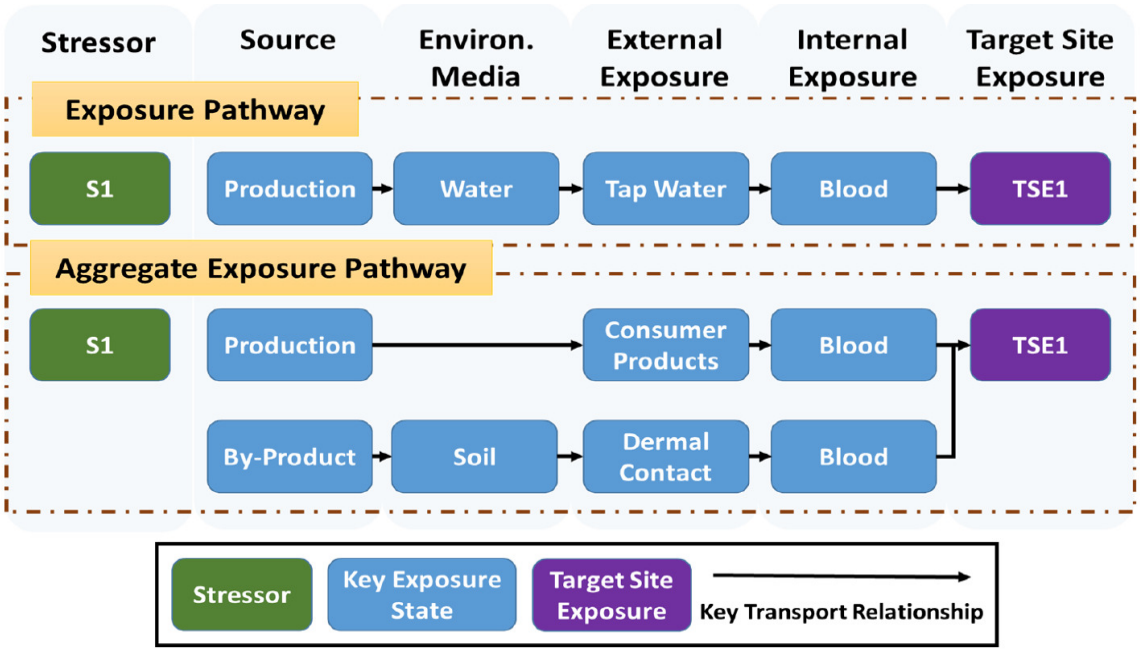



**Figure 3. Exposure Pathway and Aggregate Exposure Pathway**

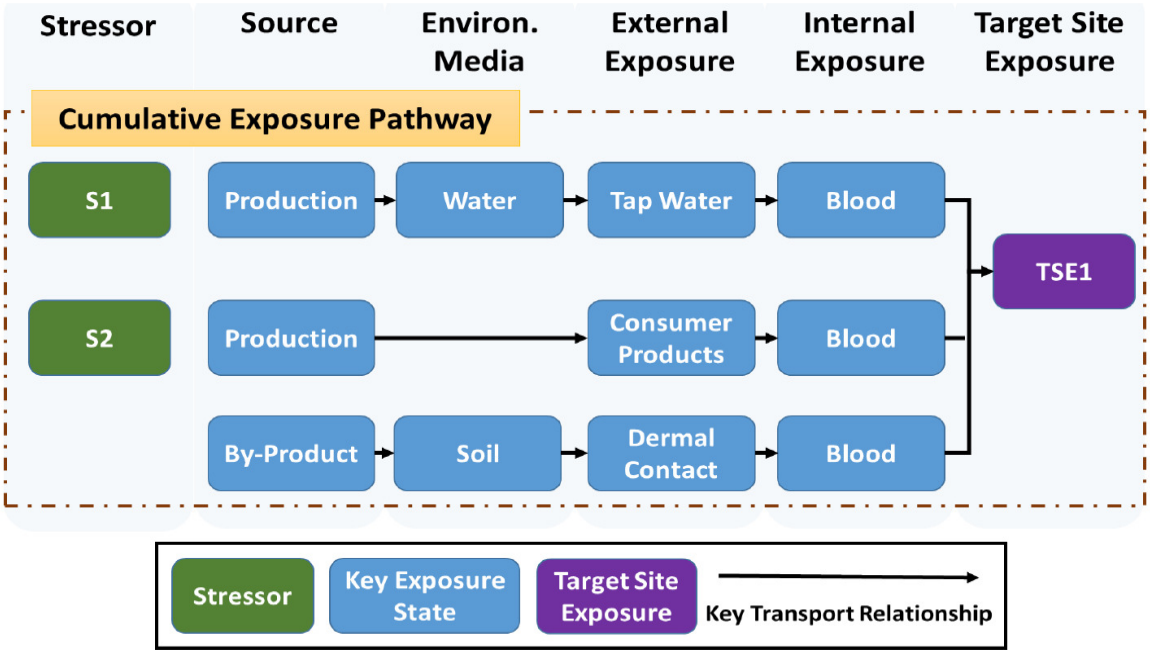



**Figure 4. Cumulative Exposure Pathway**

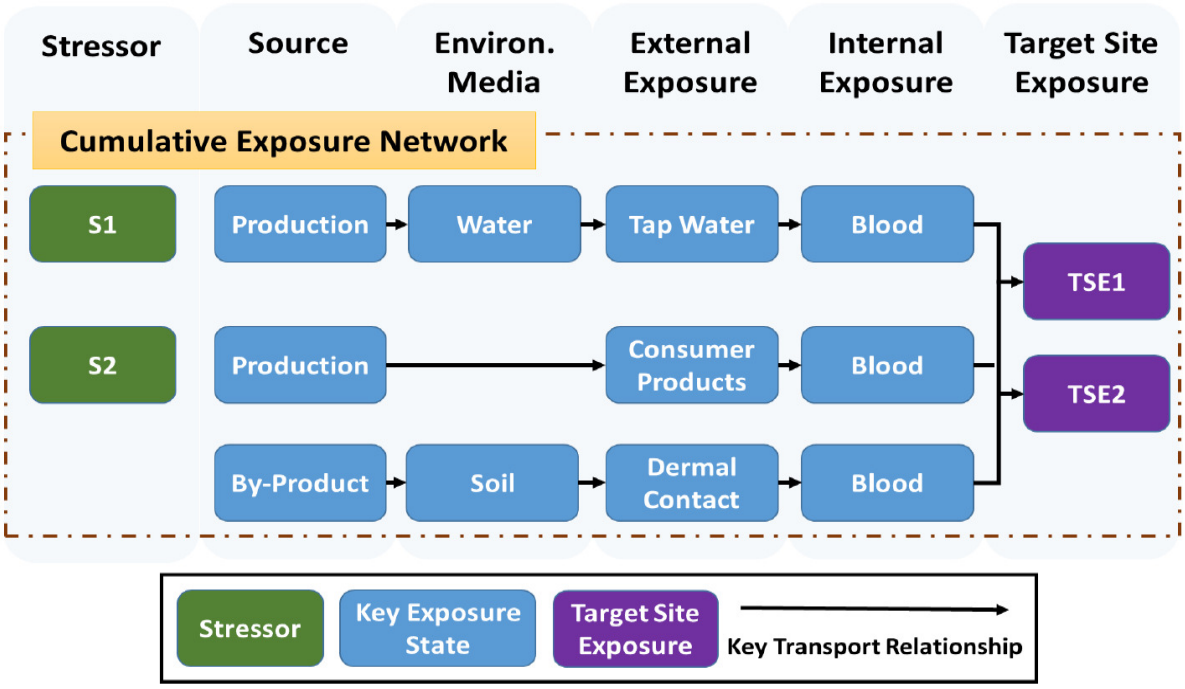



**Figure 5. Cumulative Exposure Network**

## Workgroup 2 Discussion Summary

Dr. Justin Teeguarden, the moderator for workgroup 2, began this discussion by posing the first bullet point in question 1: "How would you define an AEP?" The group began by discussing the importance of being able to incorporate the vast amount of data already out there in the literature, addressing the question "What existing concepts such as an 'exposure pathway' should be considered when defining an AEP?" Of particular note was a worry that this information would be inaccessible due to the ever-changing language used to describe the pieces of the pathway. Some stressed that there should be explicit bridges built between the AEP and more traditional models, including conceptual site models and cumulative risk assessments. The group at large did understand that the AEP was not meant to replace anything, but rather to expand upon the information already available by creating linkages between the existing knowledge and future research.

The group then moved the discussion to make sure that everyone understood the intention of the AEP. The group stressed that problem formulation needed to be part of development, which Dr. Teeguarden explained was part of the flexibility of a fit-to-purpose model. The complication of intertwining biological and geospatial information was brought up again, and Dr. Teeguarden explained that this was a way to directly integrate toxicological information and exposure information. The AEP drives each field, toxicology and exposure science, to consider the other when working within the AEP framework.

In terms of defining the AEP, the group voiced the following points:

- "Aggregate" implies that the same chemical can have different sources or pathways; is that constraining?
- "Aggregate" is a term of regulatory importance, and there is a difference between "cumulative" and "aggregate"

- Could the word "systems" be encompassing enough? It implies a network of knowledge that works together
- "Aggregate" implies that there will eventually be aggregation of pathways, but more than one pathway is not guaranteed
- How do we account for an aggregation of aggregates? Are they integrated?
- "*Aggregatable exposure pathways"* was eventually decided upon as a new way to define the AEP that more accurately represented its intentions and purposes

In terms of whether a single AEP can represent multiple sources of a single chemical to multiple biological targets, the group had the following questions or points:

- What does a 'single AEP' even mean? It has aggregate or aggregatable built into the title – can one exist on its own?
- Is there a way other than building from the bottom up that can lead to an eventual endpoint?
- Modularity will allow users to focus on the shared nodes of pathways between chemicals and biological targets.
- Focus must not be on modeling or collecting data on everything, but rather on relevant routes of exposure, life stage-specific KEs, as well as species-specific pathways or endpoints.
    - Should a subscript be added to the nomenclature to differentiate between these different possible pathways?

A question was raised regarding the possibility of unintentionally biasing future discoveries by only filling in information regarding what we know; this worry was assuaged by Dr. Teeguarden, who stated that if people are collecting information relevant to any pathway, it should be filled in to the AEP, which will avoid possible biases.

In regards to how you would define multiple AEPs that share the same KE (e.g., a non-specific metabolite generated from exposures to multiple chemicals), the group decided that AEPs can converge on AOPs at many different points. This includes affecting the same adverse outcome through different receptors or different molecular initiating events. Since any stage in the AEP could be a molecular initiating event, the group discussed that a secondary definition with better communication components would eventually be necessary.

The group then moved to address whether all AEPs should have the same types of KEs (i.e., source, environmental medium, external exposure, internal exposure, and target site exposure). Dr. Teeguarden explained that the purpose was to investigate if defining key exposures or key exposure relationships explicitly would make the AEP framework more or less constraining. Many of the modelers in the group began with backing up to describe exactly what the purpose of a KE or KER was: a descriptor for how the stressor moves through the environment, externally or internally. The fit-for-purpose nature of the AEP framework was stressed as some worried that scale of exposure would need to be taken into account in order to use the framework well. There was discussion for a need of separation of transformation and transportation within the KE definition to ensure that appropriate specificity was captured by the framework without constraining.

The following points were made regarding the definition of KEs and KERs:

- "Measurable" could lead to loopholes – limits to quantification with any instrumentation, which technically makes things *not* measureable.

- "Change" could also be misconstrued – change in concentration versus change as a measure of flux.
- How do we address if several events lead to a KE?
- Where do chemical properties come in to either KEs or KERs?
- Should KE be changed to "exposure event" in order to shift the emphasis to exposure? What does this then mean for biological events?
- KE could be changed to KES: the state of a stressor defined in space and time.
- KES could be changed to key transport/transformation relationship (KTR): a qualitative or quantitative description of the process that controls movement of a stressor from one state (KES) to another

The discussion returned to how changing the language at all would possibly lead to missing information that was produced long ago. This conversation was addressed by numerous people who explained that language is constantly evolving due to its dynamic nature and that the focus should be shifted to ensuring interoperability between new models and new software tools, though domain scientists in the area would also be contacted to discuss how to best replace language. In discussing non-chemical stressors, temporality of exposures was stressed

Dr. Teeguarden then described how the AEP borrows quite a bit from the AOP, a mature framework that is similar in many ways. Should the same 5 core principles from the AOP be used in the AEP? If these five core principles do not stand, which should be used? Would any of these principles pose an undue burden to an application community such as ecological health or epidemiology? The group discussed the following points:

- Problem formulation should drive the process of creating AEPs.
- Can an AEP be chemical agnostic if it also takes into account parameters such as PBPK models, fate and transport models, QSAR (Quantitative structure–activity relationship), etc.? Since we have already found that investigating each chemical individually is not productive, then how do we generalize it for applicability? Particular attention must be paid to the definition of the domain of applicability.
- Should be able to assemble these networks across media and targets; exposure community must be aware that generalizing is necessary.
- AOPs rely on weight of evidence to build; starting with what we know is where we must begin, but it will build from there.
- The AEP and AOP distinction is clear: the information we're aggregating/curating with AEPs is chemical specific, but structure and use can be chemical agnostic.
- No model does everything, and this one is no different. We must be comfortable with omitting certain things, or with only building small parts of the pathway(s) when constructing AEPs. How do we balance having the ability to explain, for example, 98% of the variation that occurs in the model while still maintaining enough integrity for use in the regulatory arena?
- A question was raised regarding why the AEP was even being used. If AOPs were created to address the explosion in microbiological knowledge, is AEP to be used to help us understand chemical toxicity? Exposure scientists understand fate and transport, so are we just trying to interface information with toxicologists?
    - This neglects greater point of being able to use the AEP for forensics for new chemicals; provisional pathways are necessary.

For the Workgroup 2 report out to the larger workshop audience, Workgroup 2, Dr. Teeguarden discussed a few of the definition refinements that occurred in workgroup 2 after further refinement with the other workshop leaders. Many of these were related to the temporal-spatial concept of many of these concepts and reflecting this nature in the definitions. We move away from a KE to a KES: The state of a stressor defined in space and time (credit to Jon Arnot). Jon is right that these elements are the only ones necessary to defining KES. KERs were changed to KTR: a qualitative or quantitative description of the process that control movement of a stressor from one state (KES) to another.

After the workgroup, KTR was further refined to "Key Transport Relationship", maintaining the definition seen above, and Transformation Relationship (TR) was identified as a qualitative or quantitative process that controls transformation of a stressor from one stressor to another. Additional definitions that were refined included source: an entity or action that releases to the environment or imposes on the environment a chemical, biological, or physical stressor(s) (taken from the EPA Guidance of Cumulative Risk Assessment Part 1, Planning and Scoping). Further, Target Site Exposure was defined as the contact between a stressor and an internal target site (e.g. target tissue, target receptor).

### Workgroup 3 Discussion Summary

Many suggestions emerged on how to define an AEP:

- A conceptual model that defines linkages from source to fate.
- All routes one chemical can take to a specific receptor through all relevant compartments.
- The path from external to internal dose.
- A series of causally-related steps that defines certainty that an event will lead to a hazard.
- Mass balance flows between compartments with factors that affect flows between compartments.

The group struggled with defining this pathway because it depends on what it would be used for. The terminology is important, but "aggregate" seemed confusing. Was it necessary of an AEP as an organizing framework to journey all the way to the target site exposure? The group agreed it was ideal to predict concentration and prioritize if something will link to an AOP. It was then questioned whether the AEP was being developed to determine internal concentration to eventually link to an AOP. While it was suggested to change "target site exposure" to "internal dose," controversy arose as this approach leaves gaps. The same framework may be used to complete partial AEPs.

Would isolating each pathway be helpful? The AOP strategy is to first connect one particular source to one molecular initiating event to simplify and convert to networks later. This approach seemed counterproductive to some participants for the AEP construct; the overall framework should be created and then the user should decide where to focus and measure. While still at odds, the group agreed the process for defining the pathway depended on the problem formulation.

The usefulness of the AEP is it brings exposure into the same realm as toxicology. This emphasized the importance of consistent terminology. The phrase "KEs" proved not useful to the group as they suggested the phrase "key determinants" instead. "Key determinants" implies how we define the pathway/model and make the construct work versus forcing it to be a biological construct.

If the pathway and conceptual model were defined, there would be a quantitative relationship and an internal dose. Then, the user would be able to run the model backwards to find the source attribution.

Finding those linkages and data gaps would assist in returning to the external exposure and source to better inform regulation.

The group had the following outstanding questions/issues at the end of the discussion:

- What existing concepts such as "exposure pathway" should be considered when defining an AEP?
- Should AEPs for non-chemical stressors or "endogenous exposures" present different considerations versus chemical stressors?
- Should all AEPs have the same types of KEs (e.g., source, environmental medium, external exposure, internal exposure, and target site exposure)?

Additionally, during the Workgroup 3 report out to the larger workshop audience, Dr. Peter Egeghy raised the following points/questions:

- Causality: is there causality at all?
- What does "key" mean? Is it necessary? Is it the most important?
- Is the direction of the arrows important?
    - The discussion regarding the direction of the arrows: if the goal is regulatory risk assessment, the arrows may need to point the other direction
- Is the goal to have an AEP link to an AOP?
- Implementation: do we want a series of very simple pathways that can be aggregated?

## Panel Style Group Discussion and Discussion Regarding "Definition and Development of the AEP"

*Led by Monica Linnenbrink | U.S. EPA ORD/IOAA*

Monica Linnenbrink opened up the floor for discussion amongst the workshop participants following the breakout report backs. In general, participants felt that the framework needed to include more description and use terminology that does not drift too far from the lexicon currently used by the risk assessment and exposure communities.

Specifically, workshop participants provided the following comments regarding the nomenclature used and components presented currently in the draft AEP framework:

- **Peter Egeghy (EPA/ORD/NERL):** Each group had problems with the word aggregate being used in the name of the framework.
    - **Justin Teeguarden (PNNL):** In simplest form, the framework represents an exposure pathway.
    - **Rose Zaleski (ExxonMobil Biomedical Sciences):** The AEP framework is a great concept. It helps with organization and integration. Definitions and words are really important. One chemical, many sources. Maybe call it assembled or applicable exposure pathway?
    - **Annie Jarabek (EPA/ORD/HHRA):** I agree with Rose Zaleski. I do think that though we talked about bridging current methods, some are talking about separating cumulative exposure to get at multiple stressors.
    - **Ron Hines (EPA/ORD/NHEERL):** Maybe, aggregatable would be better?
- **Stan Barone (EPA/OPPT):** Maybe there are multiple paths/multiple stressors that go into a pathway, not just multiple routes. One thing we discussed in Workgroup 3 was the meaning of

the arrows from modeling perspective – arrows are what you would parameterize vs. boxes are what you would estimate.

- **Vasu Kilaru (EPA/ORD/NERL):** One issue is that the AEP is overall framework, and once some branding has happened, hard to go away from it. But maybe ensemble exposure pathways. Don't always get to go from source to target site. It depends on what you are trying to do and don't always have totality of information, so often assembling parts.
- **Justin Teeguarden (PNNL):** Annie Jarabek and Judy LaKind have been constructive in their warning to us that there is already many years of experience in other fields that have identified language, and if we can use/adopt what is already there, that would be better. Aggregate is one of those words that has a long history of use. If we are going to make a change, we are going to have to justify and communicate why we diverge from existing terms.
- **Stan Barone (EPA/OPPT):** I brought this issue up about creating new terms in Workgroup 3. We already have exposure models and parameterized names in many of them, so why would we try to be consistent with AOPs?
    - **Justin Teeguarden (PNNL):** We want to drift as little as we have to. That terminology is understood well by the modeling community and want to maintain as much as possible.
    - **Stan Barone (EPA/OPPT):** Also, there is risk assessment lexicon. When talking about ontologies, need to consider risk assessment terminologies since people in this room are not really risk assessors.
    - **Annie Jarabek (EPA/ORD/HHRA):** In terms of multistressor, would really advocate for using cumulative.
    - **Justin Teeguarden (PNNL):** It is a difference of where we start and where we end up. We might start with exposure pathways that then get aggregated and then as you assemble, get more at multistressor.
- **Stephen Edwards (EPA/ORD/NHEERL):** We are trying to figure out use of this – research tool or regulatory tool, and we want both. When we start talking about nomenclature associated with the modeling, on AOP side, models tend to be more elaborate than AOPs in practice. Having a loser coupling is better for discussion.
- **Stan Barone (EPA/OPPT):** For AOPs, there are key events and this term has been defined. For the AEP, you are talking about key parameters in modeling, and don't see those as key events per se. So, don't see the need to be wed to this. Instead of trying to press something into service to be consistent. We didn't settle on anything in Workgroup 3, but key parameter, key determinant is what we discussed instead of trying to confuse the exposure assessor.
- **Olivier Jolliet (University of Michigan):** I like "state" or KES. I wouldn't put determinant there. Key determinants are factors affecting flow (e.g., temp, behavior of people).
- **Cecilia Tan (EPA/ORD/NERL):** If we want to have generic framework, do we need to include key determinants in pathway? Also, would key determinants be the same thing across pathways (e.g., are behaviors always important) and they should be included?
- **Olivier Jolliet (University of Michigan):** Parameterizing the flows and factors influencing the pathway.
- **Annie Jarabek (EPA/ORD/HHRA):** The struggle with using the AOPs in risk assessment is the influencing parameters, or parameterizing the arrows.
- **Justin Teeguarden (PNNL):** The KTRs capture that. For example, you would say that the key transformation event is metabolism, and you would put in parameters for that (e.g., rate

constants). Someone can measure state in one box, measure state in another box and the arrow represents the relationship between the two. What I'm hearing, though is we need to pull it out as it is getting buried.

- **Dan Villeneuve (EPA/ORD/NHEERL):** In AOP framework, there is information that goes into each module. Information content for each module folds in parameters/determinants.
- **Stan Barone (EPA/OPPT):** The narrative between each box needs to be made more explicit.
- **Jon Arnot (Arnot Research & Consulting):** With regard to transformation, the existing framework is a simple diagram. There are contexts where arrows would move linearly left to right, but they would also move back to environment. There are more complicated flow diagrams that exist within this framework.
- **Judy LaKind (LaKind Associates):** For term AEP, I would propose conceptual exposure model might be another thing to call this, so we are not limiting to site specific issues.
- **Tim Frederick (EPA Region 4):** The region represents one of the users of this information. There are standard lexicon things like external exposure, which we call that a dose. What is new to us is internal dose. There may be areas where we can standardize with existing terminology.
    - **Justin Teeguarden (PNNL):** To keep it simple, the NAS defined exposure as contact between exposure and receptor. There is no dose – it is used inconsistently. We need to be consistent if talk about dose or exposure.
    - **Glenn Suter (EPA/ORD/NCEA):** Agree that exposure is the all-encompassing term, so need to keep it in the name of this framework.
    - **Annie Jarabek (EPA/ORD/HHRA):** Need to make sure that you are explicit. In toxicology, we have dose-response. So, need to clarify terminology.
- **Kathy Plotzke (Dow Corning):** Need to decide if this framework is chemical specific or not as we think through the name. In the Teeguarden et al. (2016)[1] paper, you were thinking it would be chemical specific so aggregate makes sense. However, if this is going to be broader and tie into the AOP framework, what about "exposure outcome pathway?" The determinants are going to be fate and transport and physical-chemical properties; that is what is going to determine what state it will be in and what pathway it will go down. From a framework perspective, something that is broader and cuts across multiple disciplines and integrates and bridges is better.
    - **Justin Teeguarden (PNNL):** The reason we don't want "outcome" om the name is that we don't want to tie the framework to toxicology. This should be supporting decision making using exposure information only.
    - **Kathy Plotzke (Dow Corning):** An exposure outcome is determined by those determinants. You are going to have a release, then the outcome of that exposure (e.g., gets in fish). It is an outcome pathway. Your determinants really set which way that path is going to be. Could be multiple outcomes (e.g., contaminant released in WWTP).
- **Stan Barone (EPA/OPPT):** Word "site" calls up site-specific assessments. We don't think of site per se, we are thinking about uses. Depending on your problem formulation, you can define this, but would leave site out of it.
- **John Wambaugh (EPA/ORD/NCCT):** Part of the advantage of AOPs, as a modeler, it allowed me to communicate with other modelers. Strength of AEPs is a communication tool between different types of modelers. Whatever it is called, it is a model agnostic tool.

- **David Reif (N.C. State University):** Agree. Originators don't conceive that this is a state-path model that is linear. Arrows are more than that. It is not a PBPK style where the flow is linear. The pieces are up there and they can be moved around.

- **Rory Connolly (EPA/ORD/NHEERL):** We have been working on developing a quantitative AOP, and it differs vastly from the qualitative AOP. The quantitative version is more resource/data intensive, but you get very good predictions. I would encourage that you be clear whether the AEP is quantitative or qualitative.
  - **Stan Barone (EPA/OPPT):** In OPPT, our conceptual exposure models have solid lines for things we can quantify, and dashed lines where we can't quantify but think are important. Helps us identify data gaps and provides transparency on what you know and don't' know as well as shows prioritization.
  - **Dan Villeneuve (EPA/ORD/NHEERL):** Boxes are what you want to measure/estimate KER is how you would infer from something upstream to where it is going next. You can build layers of information that help you make decisions.
  - **Ed Perkins (USACE):** We are trying to make everything too harmonized. All the different levels help in different ways and one doesn't exclude the other. There are different approaches for developing models than for the stick models you might start with. Somehow you need to cover a whole range.
  - **Justin Teeguarden (PNNL):** Everything we do is quantitative by nature. So, in end, we aspire at end to build models and ability to make quantitative decisions. However, don't want to constrain people when data is not available. If we are not driven to do a better job and quantify information.
- **Judy LaKind (LaKind Associates):** No matter what we call the framework, we will need a lot of outreach and education. Have heard that AEP would include ADME and others say not, and confusion about lots of other things. Afraid we are focusing on naming a thing that is not fully defined and if we define, maybe easier to name it.
- **Dan Vallero (EPA/ORD/NERL):** Why can't what we are talking about be the exogenous component at AOP.
- **David Balshaw (NIEHS):** In Workgroup 1, we ended up wth hierarchy of what are the layers that you are building when using the framework of the AEP to apply it in different areas. Why are we getting stuck on things that call it a pathway? It is not a pathway. It is a network. Dunk the whole pathway idea and call it a framework that looks at exposures conceptually.
- **Glenn Suter (EPA/ORD/NCEA):** Want to respond to angst about the phrase conceptual model. Frameworks for ecological risk assessment and human health risk assessment mandate that you create a conceptual model for problem formulation. People use conceptual models to guide creation of an implemented model. They use them heuristically, which seems like what you intend to use AEPs for – to communicate.
  - **Justin Teeguarden (PNNL):** Don't have a problem with it. My concern is what it communicates more broadly. People not in our field, may think it is not application ready yet.
  - **Stan Barone (EPA/OPPT):** Don't see issue with conceptual. We are coming up with an aggregated approach and conceptualizing how this will happen, is another thing. How many AOPs are used in risk assessment? This is going to have value regardless if linked to an AOP.

- **Cecilia Tan (EPA/ORD/NERL):** Think we need to go back to what Judy said about defining the concept. Are we happy with the four different components as the framework?
    - **Olivier Jolliet (University of Michigan):** We need to discuss this concept further. Think boxes should be concentrations and the arrows flows. Need to be careful what we put there. Need to think about how we name the boxes and the flows at this level. Basically happy, but will prepare 1-2 slides to present tomorrow that define states and flows a little more.
    - **Stephen Edwards (EPA/ORD/NHEERL):** If you look at all AOP slides, it is linear from one end to another, but in reality, there is feedback and circular flow.

The group decided to continue the discussion on Day 2 and see where the workgroups ended up at the end of the day. Throughout the discussion, however, the workshop participants identified alternative names for the framework that they voted on to conclude Day 1. The number of votes for each name are as follows (listed in order of highest to lowest):

- Integrated Exposure Framework (23 votes)
- Aggregate Exposure Pathway (10 votes)
- Conceptual Exposure Model (6 votes)
- Exposure Pathway (5 votes)
- Annotated Exposure Pathway (2 votes)
- Exposure Outcome Pathway (1 vote)
- Aggregatable Exposure Pathway (1 vote)
- Assembled Exposure Pathway (1 vote)
- Applicable Exposure Pathway (0 votes)
- Accumulated Exposure Pathway (0 votes)
- Conceptual Exposure Pathway (0 votes)

# May 10, 2016

## What NIEHS Brings to the AEP Framework

*Presented by David Balshaw | NIEHS*

Dr. David Balshaw from the Exposure, Response, and Technology Branch (ERTB) of the National Institute of Environmental Health Sciences (NIEHS) presented information on what NIEHS could bring to the AEP framework. For the last 10 years, NIEHS has been trying to develop the ability to think about exposure in the real-world context. Dr. Balshaw mentioned the complexity of exposures and the body; NIEHS is trying to embrace these complexities and broaden the traditional views of exposure science. The NIEHS has the following three things to bring to the AEP framework:

- **Conceptualization**: The NIEHS has a lot of experience developing new concepts, and this experience and mentality will help in the development of the AEP framework.
- **Technology and Infrastructure**: NIEHS has spent over $150 million in recent years on exposure technology, so these resources could be helpful in the development of the framework.
- **Data**: The NIEHS has volumes of data which could be used in the AEP framework.

Dr. Balshaw presented the idea that the AEP framework includes exposome concepts. One of the struggles faced with the exposome concept is the incorporation of the external environment in the exposome. Thus

far, metabolites and biomonitoring has been used to represent the internal environment. Recently, however, there has been an interest in understanding the exposure through the external environment and the AEP framework is a way to look at the external environment. Dr. Balshaw discussed the three components more in depth.

An example of conceptualization is the exposome. The NIEHS does not have a specific definition of the exposome, but there are three commonalities amongst other's definitions:

- **Measurement**: It is not possible to measure everything or the totality of all exposures (multigenerational and from birth to death). However, it is possible to measure a lot of things, so there should be an aim to measure as much as possible including multiple stressors.
- **Multi-scale integration**: These include multiple time points, exposure and response. The AEP framework would help with multi-scale integration, not just internal exposure but also considering external environment.
- **Identify stressors that impact health**: Data driven (untargeted) discovery. A major portion of the exposome is agnosticism and trying to figure out the unknown versus simply trying to prove or disprove hypotheses.

The biggest source of data that NIEHS can bring to the AEP framework includes all of the NIEHS funded epidemiology studies for the last 50 years. This information is available online and includes high level information including the exposures, study population, etc. This information can be found at: www.niehs.gov/research/supported.

Another source of data that NIEHS can bring to the framework includes research from the superfund centers. The NIEHS superfund centers all have some degree of focus on fate and transport. This includes looking at where the superfund contaminants are located, how they are getting into the environment, where are they moving, and how data can be gathered for the contaminants down to the individual level. The superfund program is also working on data science resources to make the data available to larger communities.

ERTB has been working on technologies to support exposure assessments. These technologies have recently been primarily focused on personal exposures and not just measuring a single chemical in time and space. The personal exposures have been integrated with information pertaining to physical activity, behavior, use of illicit and licit substances, and other measures such as social stress to get a more comprehensive view of an individual's environment and how these interact.

NIEHS has had a focus in the past years on precision medicine and how an exposure assessment can be done on an extremely large cohort. One way to approach this situation is to create a cellphone-based approach using questionnaires. NIEHS is beginning the discussion on how to determine exposures through patient reported outcomes. Wearable sensors are also an option, but these cannot be put on all members of the cohort. Instead, the sensors would be worn by a representative population and developing modeling frameworks that can be used to extrapolate individual data to others. Satellite data and ambient monitoring data can be used to localize people in space and time to the data and then crowdsourcing would be used to help utilize the data.

Dr. Balshaw discussed the Children's Health Exposure Analysis Resource (CHEAR). This effort provides the external research community access to laboratory and data analyses that focus on children's health. The bulk of this includes a laboratory network comprised of six labs that are performing laboratory analyses on children's exposures. A data center is providing statistical support for analyses from the laboratory

studies. The coordinating center is providing internal coordination and a public interface. CHEAR is focused on targeted analyses, but this is being coupled with nontargeted analyses for discovery including response assays.

### Q&A/Comments

- **Justin Teeguarden (PNNL):** I want to provide some clarification as to why the attendees were there and clarify some of the issues that were discussed in previous conversations. Conversations prior to the workshop and questions raised show that people still think that there are great opportunities to better organize exposure data as a whole and deliver it. If that is the case, the next question is that the real point of the meeting is to not rebrand things that have been done in the past; what they are attempting is much larger. The modeling community has done a great job to create conceptual models, and this is not the only community we are trying to reach. They are trying to reach out to other people in the exposure science community that do not necessarily think like modelers. For example, toxicologists have a framework on how to do experiments and present data, but the purpose is to create a framework to reach out to the entire community with a natural way to think about the data and have an integration between those in the exposure science community.

## Integrated Exposure Framework: Examples

*Olivier Jolliet | University of Michigan, School of Public Health*

Dr. Olivier Jolliet presented a summary of how he would define an AEP. He defined an AEP as a description of the pathways that links one or several compartments of entry (source) to one or several receptors (targets). The KES is the concentration in a compartment and the KTR are the flows and addition. Key determinants/modifiers are things that affect the flows.

Figure  shows the near and far field environmental compartments.

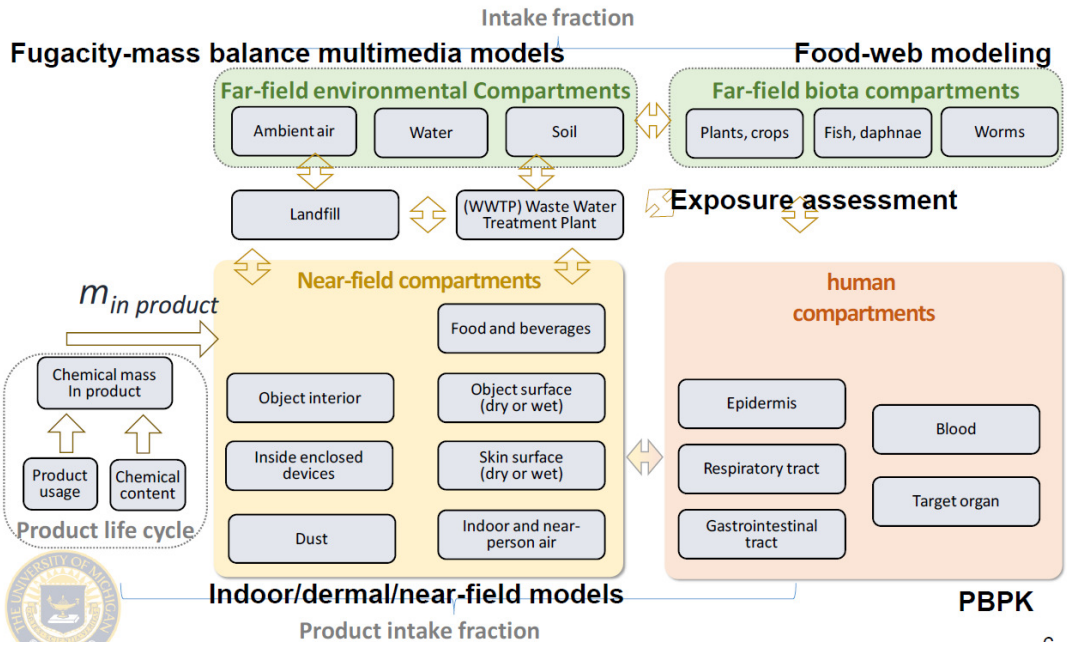



**Figure 6. Near and Far Field Environmental Compartments**

Figure 7 is an example of the AEP from compartment of entry to target receptor.

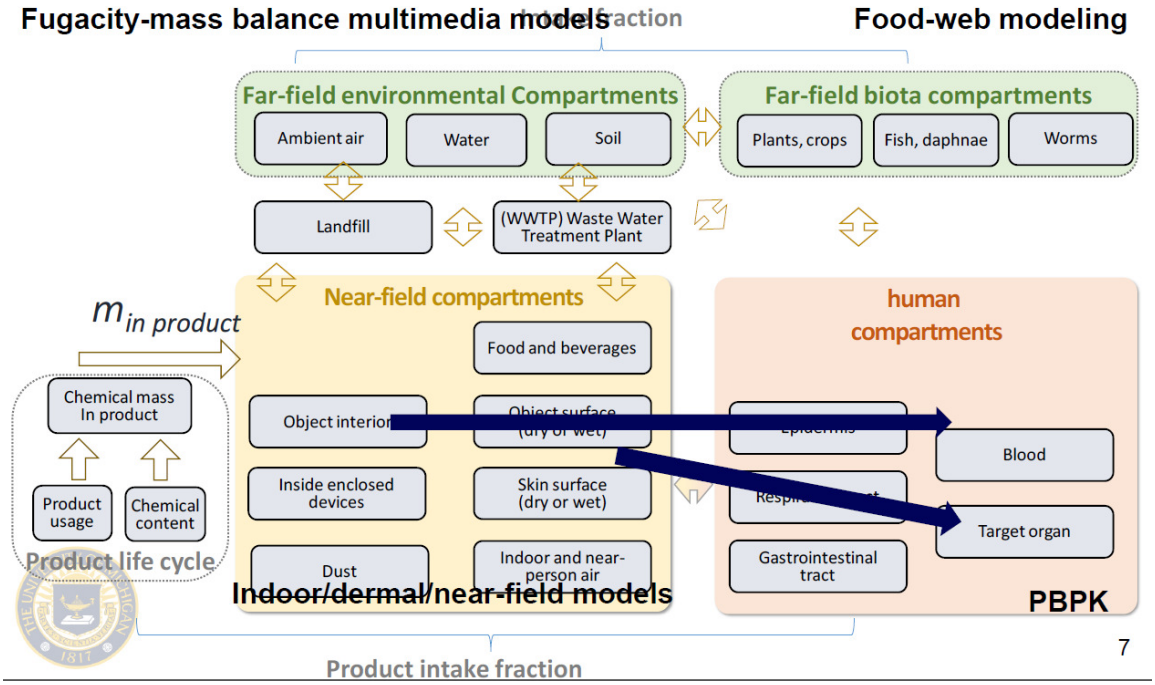



**Figure 7. AEP from Compartment of Entry to Target Receptor**

## Reprise "Definition and Development of the AEP"

*Presented by Cecilia Tan | U.S. EPA ORD/NERL*

Dr. Cecilia Tan summarized the discussions from day one to determine if there was a common ground to move onto the discussions for the second day. Definitions discussed include:

- **Exposure Pathway**: 1 source → 1 target site exposure
- **Aggregated Exposure Pathways**: multi-source, single chemical → 1 target site exposure
- **Cumulative Exposure Pathways**: multi-source, multi-chemical → 1 target site exposure
- **Cumulative Exposure Network**: multi-source, multi-stressor → multi-target site exposure
- **Key exposure State (KES):** the state of a stressor defined in space (compartment) and time
- **Key Transport Relationship (KTR):** a qualitative or quantitative description of the processes that control movement of a stressor from one state (KES) to another
- **Transformation:** a qualitative or quantitative description of the process that changes a chemical from one to another either inside or outside of the body

There was also discussion about why they want target site exposure and not just end at internal exposure.

- Target site exposure is the contact between the stressor and something else; this something else is a target exposure site. It can be a tissue or some other site.
- **Justin Teeguarden (PNNL)** clarified that it does not have to be an internal exposure solely; it can be external exposure as well. He suggested they drop the idea of it only ending in an internal site.

- The goal is to have an AEP general enough to potentially be chemical agnostic as well as chemical-specific.

## Q&A/Comments

- **Annie Jarabek (EPA/ORD/HHRA):** Just a concept to be consistent with cumulative risk assessment, the stressors may influence the arrows between the boxes. The multiple stressors may influence the arrows.
    - **Cecilia Tan (EPA/ORD/NERL):** We are trying to keep it simple, so not everything was included.
- **Oliver Jolliet (University of Michigan):** Can the name *cumulative exposure network* be used for one source/multiple targets?
    - **Cecilia Tan (EPA/ORD/NERL):** The chemicals do end somewhere and can end at different molecular initiating events.
    - **Oliver Jolliet (University of Michigan):** Some stressors can end in different people. How is this addressed?
    - **Shannon Bell (NIEHS/NTP/NICEATM):** There is a difference between "may" and "must." An exposure pathway may only have one of each because it is a single path if you have only one stressor. If you have one stressor and branch it out to different target sites, it would still be called a network. Networks may have multiples of these things, but aggregates need to be focused on one because it is focusing on a single target site.
- **Annie Jarabek (EPA/ORD/HHRA)** asked for a clarification of transport versus transformation.
    - **Cecilia Tan (EPA/ORD/NERL):** Transport is the movement and transformation is the change from one chemical to another chemical.
    - **Annie Jarabek (EPA/ORD/HHRA):** Would this change the pathways? If the nature of the chemical changes; it can become a source for another pathway.
    - **Cecilia Tan (EPA/ORD/NERL):** Transport: in the same pathways; transformation could go to another pathway; networks are connected.
- **Michelle Heacock (NIEHS):** Pertaining to external exposures, do we not have to go all the way to internal exposures? I'm confused about how this would be linked to an AOP if that is the purpose?
    - **Justin Teeguarden (PNNL):** The goal is not to use this framework to drive to internal concentration; the purpose is to organize the data. It depends on the problem that is trying to be solved.
- **Glenn Suter (EPA/ORD/NCEA):** There is not a right or wrong answer as to where exposure starts and ends; there is a different culture in the different studies. Some think of exposure as what comes into contact with the source (this is from working on PBPK models). The biggest issue is communication and the only way to reach consensus about terminology is for people to talk to each other.
- **Elaine Cohen Hubal (EPA/ORD):** The AEP is very problem-driven and this is why problem formulation keeps popping. For the AEP to add value to what has been years of work, the key thing that needs to happen is that the whole framework needs to focus on what is generalizable and what is chemical agnostic.
    - **Hristo Aladjov (Effectopedia):** When we talk about quantitative measurements for a chemical, it should be specific to a chemical; when talking about models that can handle different types of chemicals, they can be more generalizable.
    - **Cecilia Tan (EPA/ORD/NERL):** We all agree it can sometimes be stressor specific and sometimes it can be less specific**.**

- **Elaine Cohen Hubal (EPA/ORD):** In AOPs, the biology is chemical specific, but other times it is chemical agnostic. Here, the physics is going to be chemical agnostic and when you use it based on case examples, it will be chemical specific.
- **Annie Jarabek (EPA/ORD/HHRA):** It may be useful to engage stressor as a physiological factor rather than chemical specific. In the diagram that was presented, it looks like the stressor is another chemical. We may want to change the diagram to make it show that a stressor can be very generic.
- **Kathleen Plotzke (Dow Corning Corp.):** I believe it can achieve both; we can make it stressor specific and chemical agnostic. We need flexibility in the modules. Is there a way to show that you can move modules?
- **Cecilia Tan (EPA/ORD/NERL):** This is where they are trying to go with the AEP framework.
- **Nicholas Anastas (EPA/NRMRL):** Is chemical agnostic appropriate to use if we are trying to broaden the audience especially nonscientists?
- **Cecilia Tan (EPA/ORD/NERL):** They are revisiting to the names later in the afternoon.

## Concurrent Workgroup Discussions

The goal of the concurrent workgroup discussions was to think about how you would develop an AEP given the definitions provided in earlier discussions. Workgroup 1 was provided with the task to think about infrastructure, Workgroup 2 discussed applications, and Workgroup 3 talked about the human and ecological integration in the AEP framework. Summaries from each workgroup discussion are presented in the following subsections.

### Workgroup 1: Aggregate Exposure Pathway Infrastructure

One of the challenges discussed on Day 1, defining what a KES, was revisited in depth on Day 2. There was continued, constructive disagreement, but the group came to an agreement that the KE is more focused on the stressor involved, while the KER describes how the stressor moves from one state to the next, either through transfer or transformation. Data-rich and data-poor examples must be provided to make it clear how to interpret these definitions, though that was not addressed further by the workgroup. KES was determined to describe potential presence or absence of a stressor at a location. In addition, there was discussion of hierarchical systems within the AEP framework, where defining the linkages of how chemicals move from media to media will have various degrees of specificity depending on the data available at the time. As new, more relevant or reliable data becomes available, the pathways, and thus network, will grow in accuracy. From there, the group segued into a discussion on the infrastructure of the AEP.

The workgroup discussed how adoption and implementation of an AEP framework would benefit substantially from publically accessible, web-based tools similar to those developed for the AOP framework (https://aopkb.org/). These tools may contain multi-scale database and computational models. Querying a growing database of AEPs would produce new insights into the relative contributions of various sources, biological or environmental processes to total external doses, or internal concentrations at the site of action. Providing collections of modules for prediction of AEPs would provide external and internal dose estimates that can be directly linked to dose-response and hazard data for risk assessment/prioritization.

The workgroup raised the following questions that would need to be considered first before such a tool could be developed:

- **How would this infrastructure incorporate existing resources?** What existing data frameworks are candidates for inclusion? What changes in existing systems would be required to interface with the central system? How would you consolidate various exposure databases? Is there an unmet need that a new system can solve, or would effort be better spent on highlighting the availability (and possibly promoting interoperability) of existing tools?
- **Should the knowledge base include models and/or model predictions?** If so, how would you consolidate various computational models? If including model predictions, what information is required on how the predictions were made? How do we handle older versions of model predictions and ensure the content of the knowledge base is refreshed? How would we tier the information in the knowledge base to make it clear to users which data are measured and which are predicted? What should be the inclusion criteria for models/predictions?
- **How would you organize data and models (e.g., by KEs)?** What would the model/data template format(s) look like? How do we make sure that the system doesn't become limited to chemical stressors?
- **What should be the design of the new system**? What is the anticipated volume of data, analysis workflows, and queries that would be expected of the system? Is a single system or a federated system preferable? How would this system interact with existing systems (e.g. real time feeds via API, periodic updates on a release schedule)? Would it be better implemented as a catalog of existing systems? How would you design a new infrastructure for users to enter new data/information? What will be the standards for information exchange protocols and who will govern those standards?
- **How do you get buy in for such an infrastructure?** What are the uses for the KB and what implications do these have for the system design? How do we create a crowd-sourcing environment that will encourage scientists to contribute to the KB? Are there particular data types that we would want to emphasize in terms of new data collection and submission?

Dr. Stephen Edwards facilitated the discussion of incorporating existing infrastructure into the AEP framework. Much of the discussion focused on Dr. Carolyn Mattingly's discussion of the CTD database she has been developing. With the existing framework in the database, users have the ability to identify where missing data occurs, potentially guiding future research. Resolving differences in vocabulary has been difficult, but there is an iterative nature to the work. Some of the fundamental pieces of this database include identifying the stressor, exposure receptor, and exposure event from the literature. Because the nomenclature between this database and the AEP framework differs, there was discussion regarding where the KES or the target exposure site (TES) actually falls in the groups defined by Mattingly's database. In particular, urine was discussed more in-depth: is it a KES? It is a TES? Is it a biomarker, but not a KES? It is measureable, but it is not the KES; rather, it is a surrogate of exposure to better understand what is going on internally. It was particularly difficult to easily place biomarker measurements into one of the components of the AEP. Dr. Edwards concluded that further action is necessary to address states and relationships between states within the AEP framework for it to be successful.

Dr. Mattingly also described how her database was developed, stating that they started with a few papers and then had to reassess what the relationships between the measurements and mediums stated in the papers really meant for exposure. Learning how a different field, such as epidemiology or toxicology, frames their work was very important to developing an effective way to catalog the information. The workgroup discussed that building the framework would need to be additive and iterative. The workgroup

also discussed how to train people to use the framework and how to get buy-in from others and developed the following suggestions: providing students with training before graduation to facilitate buy-in and adding a training or data-entry special session over lunch to the 2016 ISES (International Society for Exposure Science) agenda/

Dr. Edwards acknowledged that building a database in an iterative fashion and acknowledging hierarchical levels of development will help users of the AEP better identify current data gaps. The discussion then moved to stakeholders, which it was determined will likely emerge as the framework is used. The populations identified as possible users included:

- Modelers would be excited about the datasets built into the framework
- Scientists could use the framework for hypothesis generation
- Small businesses may have a need for better sensor or measurement techniques to fill in data gaps
- Industry could likely use this tool to identify green alternatives, other innovations
- Non-governmental organizations (NGOs) could possibly use, but would call for full transparency on the components of the framework
- Risk assessors

In terms of risk assessors, the workgroup discussed the effectiveness of the AEP in the beginning of its development. Without sources of data (measurement or predicted), users will still be dealing with many chemicals and chemical classes where information is not readily available. In the case of consumer products in particular, there is no information available regarding what is in these products. With this lack of data, users will have to rely more heavily on predictive models and on correlations to deduce information. Although it is true that it is not necessary to know all the components of a contaminated site to be able to clean it up, it is worth keeping in mind that occurrence of a chemical is not equivalent to exposure, and the presence of an exposure is not synonymous with risk. Understanding the cascade of chemicals is critical to applying AOPs to a risk assessment framework, and some other understanding will be critical when applying the AEP framework. The workgroup also touched on issues of risk communication and proper uses of the framework.

Dr. Edwards brought the conversation back to issues regarding how to deal with sources. In particular, “source” could be defined in various ways. What was a TSE for one AEP could be analogous to the source for another. There is always an ultimate source, but because of the fit-for-purpose nature of the AEPs, the source can be a downstream event, or focus on internal or external exposures exclusively. In the AOP, the layers represent biological organization. In the AEP, a hierarchy of dimensional organization can be used to refine the pathways. Maintaining the flexibility in defining a source will allow users to identify the source most relevant to them (e.g., an industrial source, air/water/soil, a particular type of detergent, etc.), and it will allow metadata to more easily fit into the framework.

In conclusion, the discussion led to another suggestion of an alternative title for the AEP – the Comprehensive Hierarchical Exposure Framework (CHEF). Overall, the workgroup unanimously agreed that a first step for the framework was that it must build upon existing efforts and ontologies. The knowledge base should be easily accessible and incentives for use should be employed so as to encourage its development. More specific conclusions from the discussion include:

- Existing infrastructure resources that could be incorporated into the AEP framework include CTD Exposure, existing fate and transport models, existing chemical exposure models, CPCat, NEMI, YAGO geographical coordinates. To combine these databases or infrastructures, close attention

would need to be paid to nomenclature specific to each field as well as equipment ontologies for instruments.

- The AEP knowledge base should include data from model predictions. Data from model predictions should be treated similarly to how it is treated now; users must understand the quality of the predictions just as they must understand the quality of the data they use. Tiering of models should occur, perhaps based on if they are published, used in regulatory settings, peer reviewed, etc. Evidence codes, similar to those used in genomics, can be employed to help users understand how much trust should be placed in the predicted data. One suggestion put forth regarding how to tier models loaded into the AEP framework was a survey for those uploading that would automatically assign a tier to the model based on developer answers.
- Models and data should be captured in two different layers, though data should also include model predictions. A separate source box was stressed, but it should only signify the beginning of the AEP being developed, not constrained to the beginning of the stressor life cycle.
- The design of the new system should be directed by those who will use it. There will be populators of the AEP knowledge base as well as generators of new data. However, it is likely that every populator will be a user of the database. However, it was also discussed that there is a need to keep in mind who will be a target investor or supporter to avoid having the knowledge base fall to the wayside after the initial grant runs out. Keeping up that amount of data is not feasible for academia. Ensuring that standard data or axis formats are conserved throughout data entry, as well as developing the knowledge base with open source software, will considerably reduce the burden of the developers in keeping the database relevant over time.

### Workgroup 2: Applications of the Aggregate Exposure Pathway Framework

It is expected that a broad range of applications can emerge from implementation of the AEP framework within publically accessible, web-based tools such as those in development for the AOP framework (https://aopkb.org/). For example, providing modular descriptions of exposure pathways would allow for using individual modules as units of predictions (e.g., CalTox, IMPACT, USEtox), or collecting multiple modules for predicting a more complete exposure pathway. Additionally, querying a database of AEPs would produce new insights into the relative importance of key sources, such as biological or environmental processes contributing to target site exposures linked to specific AOPs. Mining of a database of AEPs would support a more efficient identification of groups of compounds acting through a common AOP, and organizing large bodies of information from exposure and toxicity studies with chemicals would inform the assessment of the exposome. To facilitate the discussion on potential applications, the workgroup considered the following questions:

- What other applications do you envision?
- Which would be the most impactful? In what area (e.g., risk assessment, source mitigation, cumulative risk, informing the assessment of the exposome, identifying data gaps)?
- Which applications can be implemented in the short term?
- Which applications would benefit the exposure science community the most near term and long term? The toxicology community? The epidemiology community?
- Which applications would pull toxicology, epidemiology, and exposure science together?
- Are there applications that are likely be used in academic research, or be the topics of research grants?
- Are there applications that would help evaluate the significance and impact of basic research in exposure science and toxicology?
- Are there example applications available now? Ecological exposure assessment?

Based on discussions, the workgroup came up with the following list of potential applications for the AEP framework:

- Risk-based screening: which ones can we be confident about moving forward? It should assimilate exposure data for prioritization.
    - This did not go all the way out to the molecular initiating event. It was useful to go to external source concertation, but in some cases we would want to go to internal concentration.
    - How do you take chemicals that are widespread and determine whether or not there is exposure?
    - Is the exposure even plausible?
- Description of potential pathways that determine whether there is a probability of exposure.
- An encyclopedia of pathways to consider for exposure.
    - Arsenic example
- Lifecycle assessment
    - Lifecycle inventory
- Comparative exposure assessment
- *In vitro* to *in vivo* extrapolation and read-across
- Increase in confidence: evaluating/interpretation of a putative KES and building weight of evidence regarding whether the KES is reasonable or an anomaly (i.e., determining whether you really believe in what was just measured).
    - Non-targeted heroin
    - Non-targeted DEET
    - AEP for support of chemical identification and dose calculation for non-targeted exposure assessment
- Agnostic application: QSAR chemical in dust, use, physical chemistry, degradation, etc. This may even include production information.
- Conceptual site model
- Addressing issues of scale
    - Time (temporal)
    - Spatial (national, personal, GIS)
- Identifying and prioritizing data gaps for research needs.
- Consumer products – down the drain-water – intermediate transport (this is a specific example of how an AEP would be useful)
- Tiered applications
    - General/conceptual
    - Qualitative
    - Semi-quantitative
    - Chemical specific (or quantitative pathways)
- Re-purposing data, standardization and best practices
- Inventory (and mining) of data/lit for each KES
- Comprehensive exposure assessment
- Allow of peer review/crowd-sourcing
    - Some hesitancy was raised to this application
- Build bridges to communities
- Access of data for toxicological/epidemiological academic community (link to other knowledge/data frameworks)

- Biomonitoring equivalence
    - Interpretation of biomonitoring data
    - Occupational
- Chemical prioritization
    - E.g., tungsten bullet
    - Other chemicals produced in the army: are they toxic to ecological?
- Generic Models
    - Applicable in the communication (creating a standard pathway information model that could possibly tie to the methods: e.g., analytical methods etc.)
    - Scenario or chemical class pathway/use class specific for groups of compounds that people could generally use
- Systems information (e.g., flow rates, temperature)
- Operationalize the conceptual model
- Organizing large bodies of information from exposure and toxicity studies with chemicals would inform the assessment of the exposome
- Mining of a database of AEPs would support a more efficient identification of groups of compounds
- Querying a database of AEPs would produce new insights into the relative importance of key sources, such as biological or environmental processes contributing to target site exposures
- Providing modular descriptions of exposure pathways would allow for using individual modules as units of predictions (e.g., CalTox, IMPACT, USEtox), or collecting multiple modules for predicting a more complete exposure pathway
- Exposure conceptualization for emerging research questions
    - Microbial multigenerational outcomes
    - Global warming
    - Prioritize research and funding

In order to get a better understanding of the applications of the AEP framework, the workgroup looked at four examples more in depth to determine how the framework would look for specific applications. The first application discussed was exposure prioritization for selection of chemical alternatives. This application included three parts: 1) using an AEP to determine if an alternative would make a significant contribution to existing exposure to a compound, 2) performing absolute exposure ranking, and 3) conducting exposure probability and hazard ranking. Example pathways were drawn for this application as shown in Figure 8.

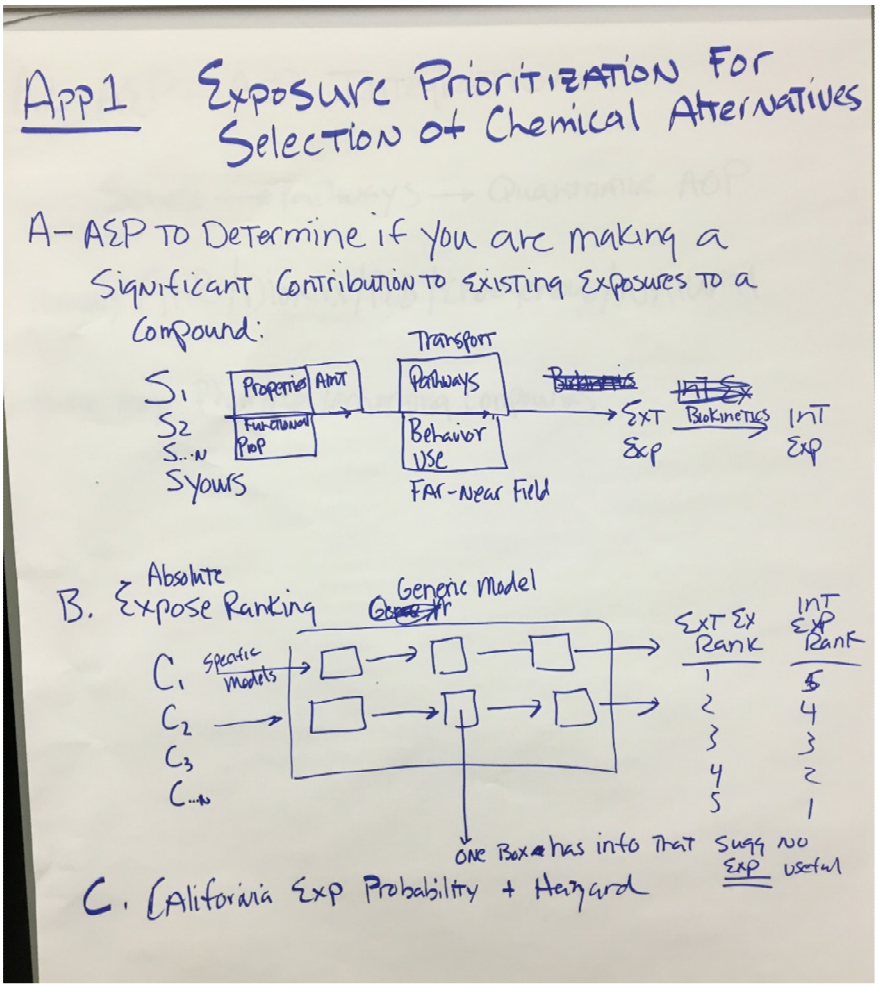



**Figure 8. Exposure Prioritization for Selection of Chemical Alternatives**

The second application explored in more detail was an AEP-AOP integration. This would include going from source to pathways and ending in a quantitative AOP. A few chemicals that would be good case studies for this application include AHR, dioxin, and PCBs. A human-ecosystem example would be pharma and other emerging compounds.

The third application explored in detail was confidence in nontargeted biological and environmental monitoring, an unknown analytical framework. For example, if a dust sample was taken, the AEP framework would be used to help match potential chemicals in the dust sample based on pathways and probabilities. This would assist in taking a sample with hundreds of chemicals and reducing the number of chemicals evaluated down to a few high probability compounds. Similar methods could be used for internal biomonitoring.

The last application explored in more depth was the use of AEP information for down selection and prioritization using a tiered AEP application. The tiered process would include: 1) initial, "crude" AEPs with available data, 2) more complete pathways with more data, 3) external to internal translation for pathway, 4) additional biological and environmental data, and 5) quantitative AEP-AOP. This tiered AEP application would be useful with applications such as ExpoCast with each tier helping to prioritize chemical information.

## Workgroup 3: Integration of Ecological and Human AEPs

The concept of the AEP was built on the long history of aggregate exposure assessment as a key feature of the field, recent advances in development of more inclusive ecological and human exposure models, and the rapid rise and application of the AOP framework. The concept of AOPs emerged from the field of ecotoxicology as a means to understand and predict potential adverse effects of chemical exposure in wildlife populations. The AOP framework has since been extended to apply in human health risk assessment. With this context, during the first half of the breakout session, the workgroup discussed the importance of AEPs being complementary to AOPs. The purpose of AOPs is to organize knowledge to identify linkages at different levels of biological organization in a causal manner.

This led to the question of whether it was necessary for the AEP to go all the way to the target site exposure/molecular initiating event. Some of the workgroup indicated that it was not necessary, and in the end depends on problem formulation ("fit for purpose") and may not need to link to an AOP. The definition might define a complete AEP, but in practice, it may be any portion of that. There is an aspirational vision to end at the target site exposure, but it may not always be the application. It doesn't always have to be complete, but it has the flexibility to account for different activities. While being able to restrict the number of columns helps with organization, the number of columns should also have the ability to expand to remain flexible.

The second argument was that there were compelling reasons to map it all the way to the molecular initiating event: 1) as a collaborative effort as other users may have an interest in other parts of the pathway, and 2) laying the framework out conceptually would help bridge gaps. The group agreed there was a need for a larger framework, but the pathway ultimately depended upon problem formulation. Finally, a large discussion unfolded on whether a single AEP can have multiple receptors and multiple target sites. To help facilitate the discussion, the group developed criteria (see table below) for choosing an example case study to work through.

**Table 1. Criteria for Case Study Example (Brainstorming Exercise)**

| | |
|---|---|
| Existing risk assessment – modify to fit within framework | Near field or far field exposure or both |
| Applicable to both human and ecological | Transformation as a key transformation relationship |
| Data rich | Simple |
| Data poor – include lumping/generic models based on chemical properties | Complex |
| With and without the ability to utilize predictive models | Multiple target sites for different species/organisms |
| Read across for exposure assessment | Product mixture (single product/source with multiple chemicals) |
| Multiple potential source of exposure (e.g., eco and drinking water) | Screening/prioritization |
| Bioaccumulative chemicals/potential human consumption issues + direct effects (PBT) | Quantitative risk assessment |
| Well-developed AOP | Lifestage issues |
| Measurements in multiple media (environmental media and biomonitoring in humans) – what are the risks and exposures? There are gaps in the path. | Existing RA – modify to fit within this new framework |
| Complete exposure pathway | Applicable to both human and eco |
| Incomplete exposure pathway (intentionally stopped) | |

Dr. Stan Barone presented the conceptual model for hexabromocyclododecane (HBCD) as a case study example (see Figure 9) because it met several of the criteria listed in Table 1: it is persistent, cumulative, and toxic; it affects ecological and human health receptors; some of its effects are linked to thyroid hormone disruption, which has an AOP; there is potential for near field and far field exposure. The molecular weight is heavy and persistent in environmental media and will partition less in water column and more in sediments.

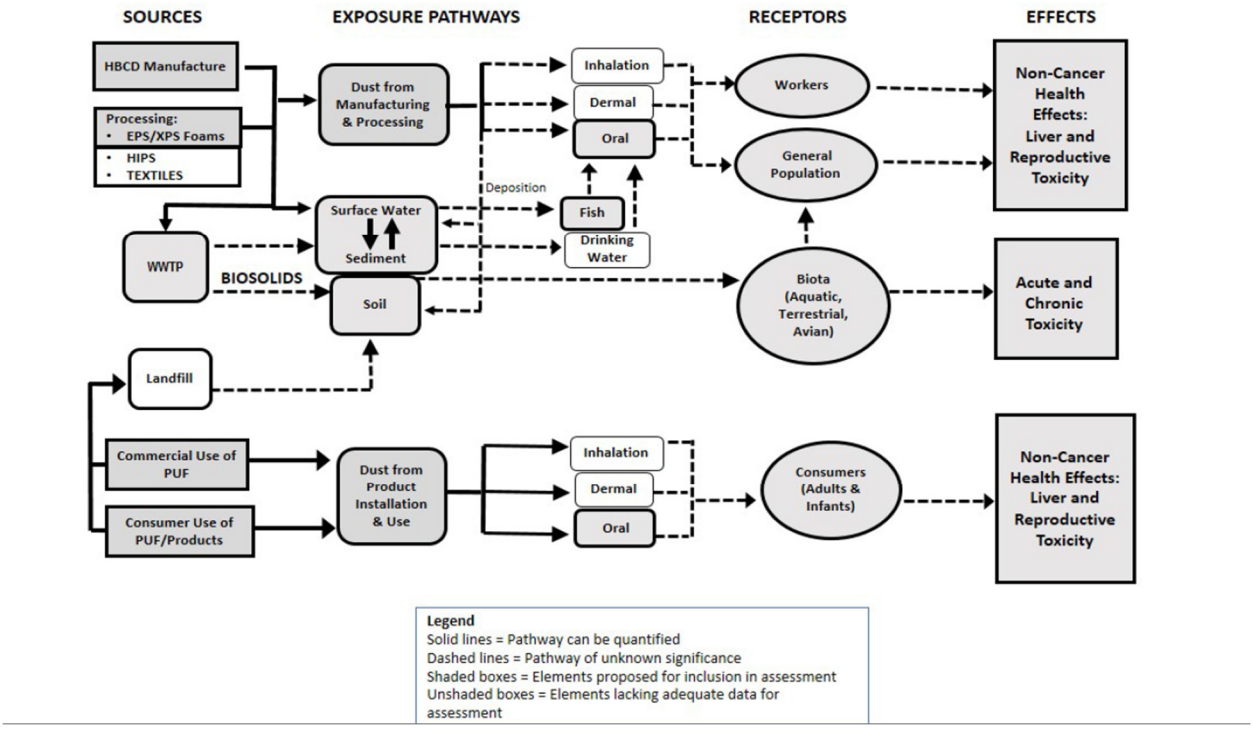



**Figure 9. Conceptual Model from Problem Formulation: Example from HBCD Assessment**

In the world of toxics and industrial chemicals, EPA has the authority to deal with, under the Toxic Substances Control Act, the manufacturing, processing, disposal, and consumer uses of chemicals, but not so much the legacy contamination. In the conceptual model, for the manufacturing process, solid lines mean more characterization and it may be possible to estimate and quantify. Dashed lines means there is a pathway but don't have enough information to estimate. The primary route for manufacturing is construction materials and manufacturing of homes. Release of this chemical will be house or office dust. There is monitoring data in multiple species and biomonitoring data in multiple tissues including breast milk, blood, fat, etc. Populations of concern include workers, general population, biota, and consumers. This chemical will be banned and phased out over the next couple years. It has a lot of data and is also missing a lot as well. EPA makes decisions in the absence of information. Refinements and improvements with a conceptual model is a good thing. It is a different structure than from what was presented. This conceptual model allowed the group to determine how to move forward in developing and implementing the AEP.

Dr. Tina Bahadori mentioned she thought the conceptual model approach is the harder approach. Instead, she suggested that it might be easier to take a case study, perform a literature search for the data, and let the data do the talking. She argued the purpose of the AEP exercise was the aggregation of data, not

knowledge. For the majority of case studies, there won't be much data available, and the majority of the boxes would be empty. Dr. Michelle Embry explained even if they didn't have the data, they could use the boxes for other purposes.

The workgroup used these thoughts and suggestions to address the following charge questions.

- Would ecological AEPs and human AEPs have the same definitions, or is there a need to define ecological AEPs separately?
    - "Receptors" is a completely different issue for ecosystems (goes far beyond a limited number of "test species")
    - The "definition" can be the same, but they would be implemented differently.
    - They aren't separate, but an extra layer is needed to define receptors.
- Would ecological AEPs and human AEPs have the same applications?
    - Yes, risk assessment is an example.
- Under what conditions does it make sense to separate ecological and human AEPs?
    - Yes, if an exposure is not relevant to humans. Yes, for something like an occupational exposure.
    - Many examples of different ones for fish than for humans (e.g., ammonia).
    - Again, they are not different conceptually, but they are different in their implementation.
- Would ecological AEPs and human AEPs have the same KEs and KERs? Do AEPs for different ecological receptors have the same KEs and KERs?
    - Receptors must be defined.
- Is there a need to separate ecological and human AEPs?
    - The framework can fit both and the AEPs are similar.
    - AEPs are not separate but receptors are different.
- Does the AEP concept enhance ecological risk assessment by providing a common structural formulation of exposure relationships and pathways that is relevant and transferable to all/most ecological receptors (and between human and ecological receptors)?
    - Differences may be more important in implementation than conceptualization. Can be lumpers in conceptualization. Model implementation will be different.
    - Pathways can obviously diverge between nonhuman and human organisms (e.g., fish respire water).
- Can the ostensibly linear framework of AEP accommodate the cyclical interactions that are characteristic of ecosystems?
    - Yes, it can be captured both qualitatively or quantitatively
    - "It's all in the arrows."

Finally, the workgroup concluded the discussion by summarizing some final thoughts and identifying questions that needed to be discussed further:

- Adding another column to the AEP framework as a key receptor state would allow us to capture the receptor more clearly.
- The framework should be flexible enough to add compartments that fit to the human health or broader ecological concerns.
- The framework approach will work for both human and nonhuman, but after the first three columns, there is a need to distinguish between the two with an additional column to keep from making divergent frameworks.

- How should we address taxonomic applicability? Should there be multiple pathways or just branching?

## Report Out: Discussion on "Definition and Development of AEP"

### Workgroup 1

Dr. Stephen Edwards provided a summary of Workgroup 1's developments during Day 2 of the workshop. He acknowledged that at this point, it was likely premature to commit to a system design. More stability is needed in the underlying definition of an AEP in order to move forward. Having a proposed definition that is flexible and easily applied to several different system designs will be key. Furthermore, considering the system as a whole, per the discussed Comprehensive Hierarchical Exposure Framework (CHEF), would be more helpful in determining optimal organization and/or system design.

He also mentioned the further refinement of the KES definition, stating that KES establishes that a chemical is measurable, and that specific measurement data should be stored separately. A proposed definition for source was also set forth by the workgroup. Analogous to the AOP context, TSE and sources can exist anywhere along a continuum; each become a special type of KES. To distinguish between transformation and transportation of KES, KTR* get us from one stressor to another through transformation while KTR gets us from one media to another through transportation, as observed in Figure 0.

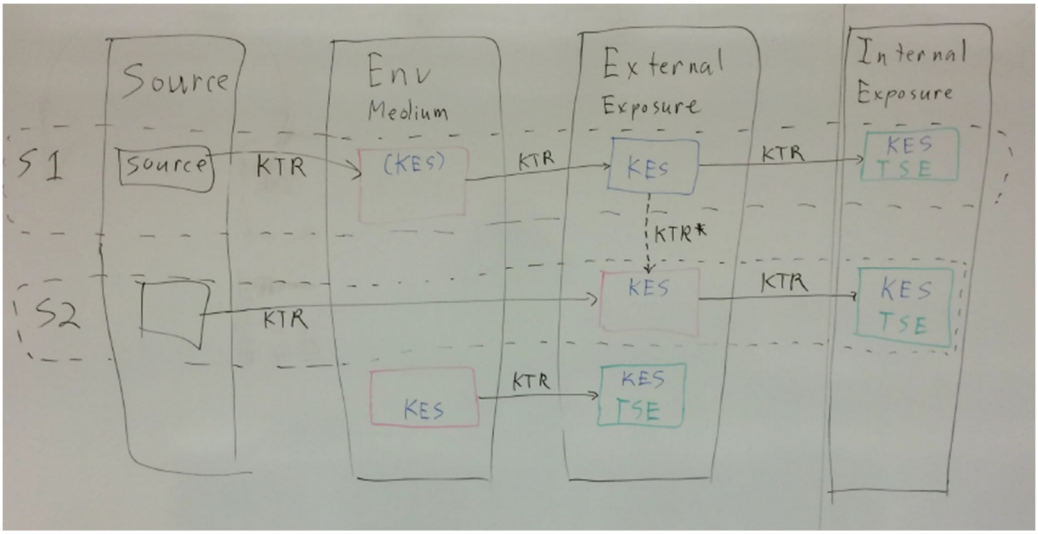



**Figure 10. Interplay between newly revised definitions of KES, TSE, KTR, and KTR*.**

Dr. Edwards summarized that modeled data would be essential. The quality of model data will require different information, but conceptually this is not far from addressing normal data quality concerns. Mechanisms are available to capture this information, and models themselves should be stored in a different layer of the AEP repository than the data. A method for organizing data and models is demonstrated pictorially in Figure 1.

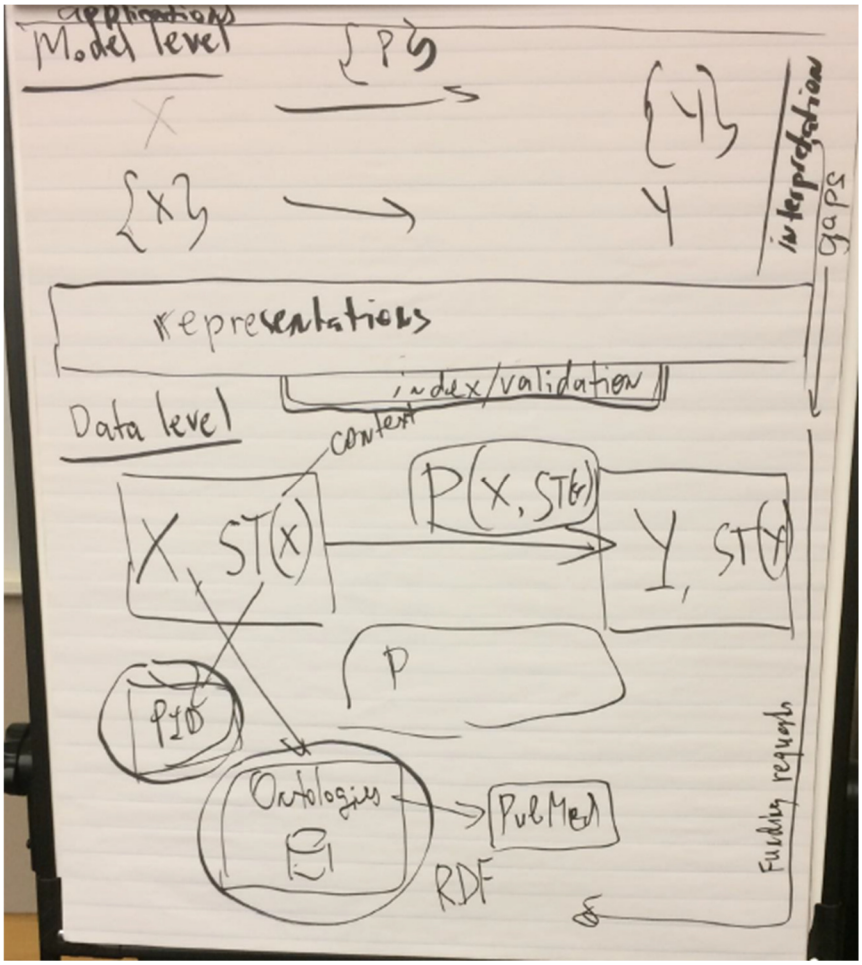



**Figure 11. Method for organizing models and data separately**

The existing resources and ontologies necessary for building the AEP repository were also mentioned. A need for harmonizing nomenclature across other options and building hierarchical structures within the AEP should be the short-term goals for this framework's development, rather than building a new system. There are many options out there for what this repository may look like, and this will undoubtedly also be influenced by the stakeholders and their buy-in. Many stakeholders were identified by this workgroup (regulators, companies, NGOs, modelers, researchers, etc.), and there will be a slight difference between those stakeholders who are producers of data and those who are consumers of data.

### Workgroup 2

Workgroup 2 suggested finding near term, useful applications for the AEP framework but have advised against building an infrastructure at this point. The group believes that user groups should be quickly engaged to create interest in the framework. Both chemical specific and agnostic properties will be possible in the framework. The group did not find any barriers with the current vision of the framework.

The AEP will be a data organization and mining tool. Eventually, it is envisioned to be a pathway information resource. Lifecycle assessment will be an important aspect of the AEP. The AEP will be useful in exposure weighing as well as for translation of exposures across test systems.

## Workgroup 3

Workgroup 3 discussed the importance of AEPs being complementary to AOPs:

- AOPs organize knowledge to identify linkages at different levels of biological organization in a causal manner
- Both identify the gaps in data/knowledge
- General agreement that goals are similar

They then discussed the question of whether or not it is necessary for the AEP to go all the way to the target site exposure/molecular initiating event:

- One argument is no; the end depends on problem formulation ("fit for purpose") and may not need to link to an AOP.
- The other argument is that there are compelling reasons to map it all the way to a molecular initiating event: 1) as a collaborative effort wherein other people may have an interest in the other parts of the pathway, and 2) laying it out conceptually will help us bridge knowledge gaps.

There was a large discussion on whether a single AEP can have multiple receptors and multiple target sites, so the group decided to work through an example and develop criteria for choosing that example (see Table 1). A HBCD flame retardant conceptual model was used as the case study example (see Figure 9). The group determined the following key issues with human and nonhuman receptors:

- Adding another column to the AEP framework as a key receptor state that would allow us to capture the receptor more clearly.
- The framework approach will work for both human and nonhuman organisms, but after the first three columns, there is a need to distinguish between the two with an additional column to keep from making divergent frameworks.
- The framework should be flexible enough to add compartments that fit to human health or broader ecological concerns.

A common theme emerged from the discussion: ecological and human AEPs can be the same in conception but different in implementation.

## Q&A/Comments

- **Stan Barone (EPA/OPPT):** Workgroup 2 had discussed the need to engage stakeholders and the community but advised against building an infrastructure just yet. There is a lot of information out there that would be applicable and helpful for the AEP framework. Did the workgroup discuss how to share this information?
    - **Justin Teeguarden (PNNL):** No, this was not discussed.
    - **Stan Barone (EPA/OPPT):** Just as a recommendation and to restate, there is a lot of information out there as far as conceptual site models and other models that could form this systematic framework that we are trying to develop and build upon. There should be a mechanism for sharing this information. We need to improve communication.
    - **Justin Teeguarden (PNNL):** This could relate to the manual of exposure pathways that was discussed in Workgroup 2. On one hand it was discussed to have this emerge versus building it initially.

- **Stephen Edwards (EPA/ORD/NHEERL):** Rather than building a system or any physical thing, it would be helpful to think about how all the pieces might fit together first and try to identify the relationships first and using the data that is already out there to show the models.
- **John Wambaugh (EPA):** Start on the ontologies efforts and start on something like GitHub.
- **Shannon Bell (NIEHS/NTP/NICEATM):** Sharing information requires a platform, and if you want to take something to a stakeholder, you need to figure out how to package it. I advise against having something built that was not originally built for the final purpose because no one is going to want to scrap something and start over.

## Appendix A. Final Agenda

**Monday, May 9, 2016 – U.S. EPA Auditorium (Room C111)**

| | |
|---|---|
| **Morning Agenda Overview:** Welcome, introductions, and a series of presentations to introduce the motivation of AEP development, concepts of AEP and AOP, and integration of human and ecological AEPs | |
| **8:00 AM** | **REGISTRATION** |
| 9:00 AM | **Welcome**<br>Jennifer Orme-Zavaleta \| *U.S. EPA ORD/NERL* |
| 9:05 AM | **Workshop Overview**<br>Monica Linnenbrink \| *U.S. EPA ORD/IOAA* |
| 9:20 AM | **Pathway-Based Predictive Approaches: Linkage Between Exposure and Effects**<br>William Benson \| *U.S. EPA ORD/NHEERL* |
| 9:35 AM | **Rethinking Exposure Science**<br>Jennifer Orme-Zavaleta \| *U.S. EPA ORD/NERL* |
| 9:50 AM | **The Case for Aggregation of Exposure Information**<br>Tina Bahadori \| *U.S. EPA ORD/IOAA/CSS* |
| **10:05 AM** | **BREAK** |
| 10:20 AM | **Introducing Aggregate Exposure Pathway (AEP)**<br>Cecilia Tan \| *U.S. EPA ORD/NERL* |
| 10:35 AM | **Lessons Learned from the Adverse Outcome Pathway (AOP)**<br>Stephen Edwards \| *U.S. EPA ORD/NHEERL* |
| 10:50 AM | **Applications of the AEP**<br>Justin Teeguarden \| *Pacific Northwest National Laboratory* |
| 11:05 AM | **Integration of Human and Ecological AEPs**<br>Peter Egeghy \| *U.S. EPA ORD/NERL* |
| 11:20 AM | **Q&A and Group Discussion**<br>Cecilia Tan \| *U.S. EPA ORD/NERL* |
| **12:00 PM** | **LUNCH BREAK** |

**Monday, May 9, 2016 – U.S. EPA Auditorium (Room C111)**

| | |
|---|---|
| **Afternoon Agenda Overview:** Three Parallel Workgroup discussions of definition and development of the AEP for public/environmental health | |
| 1:00 PM | **Parallel Workgroup Discussion on "Definition and Development of the AEP"**<br>[Break-out rooms: Group 1 (Room C111A); Group 2 (Room C111B); Group 3 (Room C111C)] |
| 2:45 PM | **Break for Participants**<br>***Group Leaders and Recorders coordinate report-outs*** |
| 3:15 PM | **Report-Out** (Auditorium, led by Group Leaders, 10 min from each group) |
| 3:45 PM | **Panel-Style Group Discussion** (Auditorium)<br>Monica Linnenbrink \| *U.S. EPA ORD/IOAA* |
| 4:45 PM | **Develop Consensus for "Definition and Development of the AEP"** (Auditorium)<br>Monica Linnenbrink \| *U.S. EPA ORD/IOAA* |
| **5:15 PM** | **ADJOURN DAY 1** |
| **6:30 PM** | **OPTIONAL DINNER** (Mez: 5410 Page Road, Durham, NC 27703; 919-941-1630) |

**Tuesday, May 10, 2016 – U.S. EPA Auditorium (Room C111)**

| | |
|---|---|
| **8:30 AM** | **SIGN-IN at REGISTRATION** |
| 9:00 AM | **What NIEHS brings to the AEP framework**<br>David Balshaw \| *NIEHS* |
| 9:10 AM | **Reprise "Definition and Development of the AEP"**<br>Cecilia Tan \| *U.S. EPA ORD/NERL* |
| 9:30 AM | **Introduce Charge Questions for Day 2** (led by Group leaders) |
| **9:40 AM** | **BREAK** |
| 10:00 AM | **Concurrent Workgroup Discussions (Break-out rooms)**<br>Group 1 topic: Infrastructure for organizing AEP knowledge and data (Room C111A)<br>Group 2 topic: Applications of AEPs (Room C111B)<br>Group 3 topic: Integration of ecological and human health AEP (Room C111C) |
| **12:00 PM** | **LUNCH BREAK** |
| 1:00 PM | **Concurrent Workgroup Discussions (Continued)** |
| 3:30 PM | **Break for Participants**<br>***Group Leaders and Recorders coordinate report-outs*** |

**Tuesday, May 10, 2016 – U.S. EPA Auditorium (Room C111)**

| | |
|---|---|
| 4:00 – 4:45 | **Report-Out on Workgroup Directions, Annotation of Outline(s)**<br>(led by Group Leaders, 15 min for each group) |
| 4:45 – 5:00 | **Next Steps**<br>Cecilia Tan \| *U.S. EPA ORD/NERL* |
| **5:00** | **ADJOURN PUBLIC MEETING** |

**Wednesday, May 11, 2016 – U.S. EPA Auditorium (Room C111) *(Invited Participants Only)***

| | |
|---|---|
| 9:00 – 12:00 | **Writing Teams (Break-Out Groups)**<br>Reconcile pre-workshop materials with Workshop outline(s)<br>Group 1: Room C112<br>Group 2: Room C114<br>Group 3: Room A105 |
| **12:00** | **ADJOURN** |

## Appendix B. Participants

| Registrant | Affiliation | Attendance |
|---|---|---|
| Hristo Aladjov | Effectopedia | In Person |
| Nicholas Anastas | U.S. EPA/NRMRL | Webinar |
| Gerald Ankley | U.S. EPA/ORD/NHEERL | In Person |
| Jon Arnot | Arnot Research & Consult. | In Person |
| Megan Avakian | MDB Inc. | In Person |
| Mark Bagley | U.S. EPA/ORD/NERL | In Person |
| Tina Bahadori | U.S. EPA/ORD/IOAA/CSS | In Person |
| David Balshaw | NIEHS | In Person |
| Stan Barone | U.S. EPA/OPPT | In Person |
| Rick Becker | ACC | In Person |
| Shannon Bell | ILS NIEHS/NTP/NICEATM | In Person |
| William Benson | U.S. EPA/ORD/NHEERL | In Person |
| Karen Blackburn | P&G | In Person |
| Susanna Blair | U.S. EPA | In Person |
| Natalie Blanton | ICF International | In Person |
| Rebecca Boyles | RTI Bioinformatics | In Person |
| Tim Buckley | U.S. EPA | In Person |
| Lyle Burgoon | USACE | In Person |
| Michael Broder | U.S. EPA | Webinar |
| Canden Byrd | ICF International | In Person |
| Weihsueh Chiu | Texas A&M University | In Person |
| Harvey Clewell | ScitoVation | In Person |
| Rory Conolly | U.S. EPA/ORD/NHEERL/ISTD | In Person |
| John Cowden | U.S. EPA/ORD/CSS | In Person |

| Registrant | Affiliation | Attendance |
|---|---|---|
| Yuxia Cui | NIEHS | In Person |
| Richard Di Giulio | Duke University | In Person |
| Stephen Edwards | U.S. EPA | In Person |
| Peter Egeghy | U.S. EPA | In Person |
| Michelle Embry | ILSI HESI | In Person |
| Neeraja Erraguntla | ACC | Webinar |
| Kellie Fay | U.S. EPA/ORD/NHEERL | In Person |
| Michael Firestone | U.S. EPA/OA/OCHP | Webinar |
| Julie Fitzpatrick | U.S. EPA/OSA | Webinar |
| Jill Franzosa | U.S. EPA/ORD/IOAA/CSS | In Person |
| Tim Frederick | U.S. EPA Region 4 | In Person |
| Kenda Freeman | NIEHS | In Person |
| Andrew Gillespie | U.S. EPA | In Person |
| Rocky Goldsmith | Independent Consultant | In Person |
| Ami Gordon | ICF International | In Person |
| Annette Guiseppi-Elie | U.S. EPA/ORD/NERL | In Person |
| Michelle Heacock | NIEHS | In Person |
| Christine Hendren | CEINT | In Person |
| Ron Hines | U.S. EPA/ORD/NHEERL | In Person |
| Elaine Cohen Hubal | U.S. EPA/ORD/NERL | In Person |
| Kristin Isaacs | U.S. EPA/ORD/NERL | In Person |
| Annie Jarabek | U.S. EPA/ORD/HHRA | In Person |
| Olivier Jolliet | University of Michigan | In Person |
| Agnes Karmaus | ILS/NICEATM | In Person |
| Vasu Kilaru | U.S. EPA/ORD/NERL | In Person |

| Registrant | Affiliation | Attendance |
|---|---|---|
| Barbara Klieforth | U.S. EPA/ORD/NCER | Webinar |
| Judy LaKind | LaKind Assoc. | In Person |
| Meredith Lassiter | U.S. EPA/NCEA | In Person |
| Jeremy Leonard | U.S. EPA/ORD/NERL | In Person |
| Jessica Levasseur | ICF International | In Person |
| Monica Linnenbrink | U.S. EPA ORD/IOAA | In Person |
| Richard Lippin | Preventative Medicine Advocate from Southampton, PA | In Person |
| Tom Long | U.S. EPA/ORD/NCEA | In Person |
| Adeline Lopez | MDB Inc. | In Person |
| Michael Loughran | U.S. EPA | Webinar |
| Carolyn Mattingly | N.C. State University | In Person |
| Gary Miller | Emory HERCULES Center | In Person |
| Cheryl Murphy | Michigan State University | In Person |
| Jennifer Orme-Zavaleta | U.S. EPA/ORD/NERL | In Person |
| Alicia Paini | European Union Joint Research Centre | Webinar |
| Ed Perkins | USACE | In Person |
| Katherine Phillips | U.S. EPA/ORD/NERL | In Person |
| Thomas Pierce | U.S. EPA/ORD/NERL | In Person |
| Kathleen Plotzke | Dow Corning Corp. | In Person |
| Kristi Pullen | NRDC | In Person |
| David Reif | N.C. State University | In Person |
| Larry Reiter | U.S. EPA Retired | In Person |
| Natalia Reyero | Jackson State University | In Person |
| Jennifer Richmond-Bryant | U.S. EPA/NCEA | In Person |
| Magdalini Sachana | OECD | In Person |

| Registrant | Affiliation | Attendance |
|---|---|---|
| R. Woodrow Setzer | U.S. EPA/ORD/NCCT | In Person |
| Lesley Skalla | MDB Inc. | In Person |
| Jon Sobus | U.S. EPA | In Person |
| Ian Sorrell | Unilever | In Person |
| Lindsay Stanek | U.S. EPA/ORD/NERL | In Person |
| Glenn Suter | U.S. ORD/NCEA | In Person |
| Cecilia Tan | U.S. EPA/ORD/NERL | In Person |
| Justin Teeguarden | PNNL | In Person |
| Tami Thomas-Burton | U.S. EPA Region 4 | In Person |
| Mike Tornero | U.S. EPA/ORD/NERL | In Person |
| Dan Vallero | U.S. EPA | In Person |
| Dan Villeneuve | U.S. EPA/ORD/NHEERL | In Person |
| John Wambaugh | U.S. EPA | In Person |
| Pai-Yei Whung | U.S. EPA/ORD/NERL | In Person |
| Meredith Williams | California Department of Toxic Substances Control | In Person |
| Eva Wong | U.S. EPA/OCSPP/OPPT | In Person |
| Miyoung Yoon | ScitoVation | In Person |
| Rosemary Zaleski | ExxonMobil Biomedical Sciences, Inc. | In Person |
| Valerie Zartarian | U.S. EPA | Webinar |
| Ilya Zaslavsky | UCSD | In Person |
| Benjamin Zukowski | U.S. EPA | Webinar |