National Toxicology Program

Board of Scientific Counselors

February 2, 2021

National Institute of Environmental Health Sciences
Research Triangle Park, NC

Summary Minutes

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1. Abbreviations and Acronyms

BSC	Board of Scientific Counselors
Carci HEI	Carcinogenicity Health Effects Innovation
CEM	Combined Exposures and Mixtures
DNA	Deoxyribonucleic acid
DNTP	Division of the National Toxicology Program
EO-CRC	Early onset colorectal cancer
EWG	Environmental Working Group
FAIR	Findable, accessible, interoperable, and reusable
IVIVE	*In vitro* to *in vivo* extrapolation
NIEHS	National Institute of Environmental Health Sciences
NTP	National Toxicology Program
PAHs	Polycyclic aromatic hydrocarbons
PFAS	Per- and polyfluoroalkyl substances

2. Attendees[1]

Board of Scientific Counselors

Chair: David Eaton, PhD, University of Washington
David Berube, PhD, North Carolina State University
Eric Blomme, DVM, PhD, AbbVie (*ad hoc*)
Weihsueh Chiu, PhD, Texas A&M University
Susan Felter, PhD, Procter & Gamble
Kathleen Gray, PhD, University of North Carolina, Chapel Hill (*ad hoc*)
Pamela Lein, PhD, University of California, Davis (*ad hoc*)
Matthew Martin, PhD, Pfizer, Inc. (*ad hoc*)
David Michaels, PhD, George Washington University
Mark Russi, MD, Yale University (*ad hoc*)
Anne Ryan, DVM, PhD, Act 5 Ventures LLC
Veena Singla, PhD, Natural Resources Defense Council (*ad hoc*)

National Institute of Environmental Health Sciences/National Toxicology Program (NIEHS/NTP) Staff

Rick Woychik

National Institute of Environmental Health Sciences/Division of the National Toxicology Program (NIEHS/DNTP) Staff

Brian Berridge
Warren Casey
Dori Germolec
Alison Harrill
Kembra Howdeshell
Jui-Hua Hsieh
Scott Masten
Elizabeth Maull
Arun Pandiri
Cynthia Rider
Sheena Scruggs
Erik Tokar
Suramya Waidyanatha
Nigel Walker
Amy Wang
Mary Wolfe

Other Federal Agency Staff

Gonçalo Gamboa da Costa, U.S. Food and Drug Administration (BSC liaison)
Elizabeth Whelan, National Institute for Occupational Safety and Health (BSC liaison)

Contract Support Staff

Katherine Duke, ICF
Jeanne Luh, ICF
Ernie Hood, Bridport Services
June Mader, GOFORWARD LLC
Blake Riley, ICF
Samantha Snow, ICF

Public Attendees

David Andrews, Environmental Working Group
Olga Naidenko, Environmental Working Group

[1]The meeting was webcast with the listed individuals attending by Zoom. NIEHS/DNTP staff are limited to those with a role at the meeting. Public attendees are limited to those presenting oral comments.

3. Introductions and Welcome

The National Toxicology Program (NTP) Board of Scientific Counselors (BSC) convened on February 2, 2021 via Zoom for identified attendees noted above and webcast for public attendees. Dr. David Eaton served as chair. Dr. Sheena Scruggs served as the Designated Federal Official.

Dr. Eaton called the meeting to order at 12:30 p.m., welcomed everyone to the meeting, and asked BSC members, Drs. Rick Woychik, Brian Berridge, Sheena Scruggs, Gonçalo Gamboa da Costa, and Elizabeth Whelan to introduce themselves. He noted that board member Dr. Susan Tilton would not be in attendance. Dr. Scruggs read the conflict-of-interest policy statement and briefed the attendees on meeting logistics.

4. Introduction to the Meeting Agenda

Dr. Berridge, Associate Director of NTP and Scientific Director of the Division of the NTP (DNTP), introduced the meeting's agenda.

He reflected upon the feedback from the December 3-4, 2020 BSC meeting, where board members were asked the following three questions in a survey:

- Was BSC engagement at the right strategic level to enable valuable input to DNTP's direction and work?
- What went well, specifically?
- What can we do better next time?

Survey responses showed that all respondents felt that the engagement met or exceeded expectations. Respondents suggested that there should be a broader discussion of the discussion questions, with more time devoted to the discussions and fewer, simpler questions. The breakout group sessions during the meeting were well-received.

Dr. Berridge described the elements of continuous improvement for BSC meetings based on the feedback received.

- Fewer, simpler discussion questions
- Lead discussants will kick off the discussion with brief comments
- Chair will facilitate discussion with aim of broader engagement
- Team members will keep video cameras on and will be prepared to contribute
- Breakout group sessions extended

He reviewed the four areas of focus in the DNTP portfolio and discussed the agenda of upcoming 2021 BSC meetings.

There were no clarifying questions from the BSC members.

5. Carcinogenicity Health Effects Innovation Program

Drs. Amy Wang and Arun Pandiri briefed the board on the Carcinogenicity Health Effects Innovation (Carci HEI) Program. Dr. Wang introduced the Program Management Team, which consisted of Drs. Amy Wang, Alison Harrill, Arun Pandiri, Dori Germolec, Erik Tokar, Warren Casey, and Ian Chan. She presented background information about the incidence and death rates associated with cancer. She noted that the Carci HEI program focuses on the contribution to cancer from environmental exposures, and that there is still room for improvement in cancer research.

Dr. Wang described DNTP's traditional approach to cancer research, which includes the two-year rodent bioassays and mechanistic investigations of a few substances at a time. She then explained that newer approaches, including the disease-focused approach and the use of more advanced tools and methods such as high-throughput screening, are also in DNTP's portfolio. DNTP has the expertise and is well positioned to take on the challenge of fostering innovation in testing and assessment and communicating key findings on environmental cancer hazards in an efficient manner.

Dr. Pandiri described the four objectives of the Carci HEI program:

- Objective 1: Discovering new approaches for cancer hazard assessment
- Objective 2: Investigating tissue-specific human cancers
- Objective 3: Relying on resources to make existing information on carcinogens Findable, Accessible, Interoperable, and Reusable (FAIR)
- Objective 4: Increasing collaborations and stakeholder engagement

Objective 1 incorporates translation, mechanisms, and prediction, all of which will complement existing programmatic approaches to cancer assessment. It includes evaluation of genetic and epigenetic alterations leading to neoplasia or uncontrolled growth of cells. Dr. Pandiri presented a summary and data update on the next-generation sequencing studies on mouse tumors and referenced partnerships and novel approaches to examine carcinogenesis.

Under Objective 2, the Carci HEI program is focusing on early onset colorectal cancers (EO-CRC), which has seen increasing incidence rates in younger demographics. He noted that the EO-CRC research aligns with the DNTP Translational Toxicology Pipeline. Research is underway to compare mutation signatures from EO-CRC patients, rodent tumors, and human and rodent colonoids. This will bridge the translational gaps across the models.

The Carci HEI program, responding to Objective 3, will leverage a variety of resources for implementing the FAIR principles. Resources will include curated data and search tools such as the Chemical Effects in Biological Systems and Integrated Chemical Environment databases, curated chemical lists, computational models such as *in vitro* to *in vivo* extrapolation (IVIVE), and quantitative structure-activity relationship models.

A new communication strategy and new approaches for carcinogenicity testing and assessment are needed to achieve Objective 4. It will be important to optimally use all communication channels. A wide variety of stakeholders are involved, from academia to industry to government.

Dr. Pandiri described the timelines involved for various Carci HEI projects.

Clarifying Questions

Dr. Eaton asked Dr. Pandiri whether the 15% of chemical-induced tumors that had mutation signatures distinct from those of spontaneous tumors were from genotoxic carcinogens or from chemicals that were thought to act in later stages and not necessarily through mutagenesis. Dr. Pandiri replied that the list he showed included both genotoxic and non-genotoxic chemicals. Since there is a wide heterogeneity in spontaneous tumors, it is not surprising that several of the chemicals have a signature similar to that of a spontaneous tumor. Dr. Eaton asked about the importance of deoxyribonucleic acid (DNA) repair processes and how they fit into the Carci HEI program. Dr. Pandiri said that DNA repair is a very important mode of action, and the program will include investigation of several adverse outcome pathways that look at the various pathways leading to cancer.

Dr. Susan Felter asked whether all the information regarding hepatocellular carcinomas was collected from the B6C3F1 mouse model, and if there would be an opportunity to look at other mouse strains, perhaps stemming from partners who have data from other strains. Dr. Pandiri cited collaborations that will explore other mouse strains, as well as rats.

Dr. David Berube asked if there is a timetable regarding the Carci HEI program's communication plans, and whether there would be a strong commitment to a data-driven communication program. Dr. Pandiri replied that there is a commitment to communication in the program, but there is no definitive timeline. Dr. Wang added that there will be an emphasis on communication and outreach. Dr. Mary Wolfe said that those elements are recognized to be very important, based on prior feedback from the board, and more specific strategies will be devised in consultation with board members. Dr. Woychik said the need to add sophistication in how concepts are brought forward and communicated outward are currently topics of intense discussion.

Dr. Eaton asked whether there is a process for researchers not currently collaborating with DNTP scientists to access tissues from the NTP Archives.[2] Dr. Pandiri replied that the tumor tissues are available for anyone to investigate. Requests can be submitted to Dr. Ronald Herbert, DNTP (herbert@niehs.nih.gov), which will be reviewed by the NTP archives use committee.

5.1. Public Comments

Dr. Olga Naidenko, vice president of science investigations from the Environmental Working Group (EWG), presented oral comments regarding the Carci HEI program. She addressed three specific areas. First, DNTP needs to develop specific implementable actions and research programs to address the disproportionate high rates of cancer seen in certain populations and to combat systemic racism. Second, DNTP should ensure a balanced research portfolio that addresses all 10 Key Characteristics of Carcinogens. While mutagenesis is well reflected in the Carci HEI program, there does not appear to be a strong research portfolio in the areas of chronic inflammation, immune system suppression, or hormonal receptor-mediated effects. Third, EWG urged DNTP to provide greater transparency regarding partners' involvement and provide opportunities for other stakeholders to participate in a manner that would bring in the public health interest community and academic research institutions.

[2] https://ntp.niehs.nih.gov/data/archives/index.html

Dr. Eaton noted that EWG also submitted written comments on this topic that were distributed to the board.

5.2. BSC Discussion

Board members were asked to consider three questions.

5.2.1. *First Question*

Consider the Problem Statement, Objectives, and Value Proposition in the Program Concept document:

- ***Group #1: Share your insights regarding whether there is clear alignment among the three. For example, do the Objectives align with the Problem Statement? Does the Value Proposition match what is being stated in the Problem Statement?***
- ***Group #2: Share your insights on the strategic fit of the overall program with DNTP's mission, goals, and capabilities.***
- ***Group #3: Share your insights on whether there is sufficient focus to deliver the intended value to stakeholders.***

Dr. Eaton asked Dr. June Mader to facilitate the exercise for board members. She read the question and said the BSC members would break into three work groups to discuss the topics listed above. Following the groups' deliberations, Dr. Mader called upon the group leaders to report on their results.

Dr. Veena Singla provided input from Group #1's discussion (see Attachment A for slides presented). The group felt there was great alignment on EO-CRC in terms of the objectives, problem statement, and value proposition. Regarding the approach and models, the group thought the *in vivo* approach was not able to encompass complexities such as the microbiome and that the rat model selected may not be reflective of EO-CRC. DNTP should be cautious about linking the *in vitro* and *in vivo* approaches. Focusing on the mutational signatures as the outcome to determine whether substances are contributing to the carcinogenic process is limited and should be expanded upon. Dr. Singla remarked that the problem statement clearly stated the problem, focus, and ultimate goals. There was, however, a disconnect between the problem statement and integrating the disproportionately impacted groups into the objectives. DNTP should focus on the details of the communication plan and timetable and build those into the entire research plan.

Dr. Anne Ryan provided input from Group #2's discussion (see Attachment B for slide presented). The group felt the Carci HEI program was a key strength of DNTP's capabilities. It was unclear based on the presentation what training opportunities were available. There was excitement over the new tools available for use including the error-corrected duplex sequencing technology and the resources available to bridge the earlier time points to the two-year carcinogenicity studies. Dr. Eaton added that this program fits with the pre-existing mission of DNTP and there is opportunity to build upon the two-year assays to include mechanistic tools and approaches into that study design.

Dr. Weihsueh Chiu provided input from Group #3's discussion (see Attachment C for slide presented). He began by agreeing with Dr. Eaton's point and thought more clarity was needed on

how this research complements the traditional DNTP bioassays. The group was unsure how intended value was defined. Dr. Chiu used the Venn diagram as an example of what certain groups of stakeholders would find valuable, with the information falling into the three categories of methods development, results, and interpretation/communication of the results. The group felt DNTP should ensure that stakeholders with less resources have the same opportunity to access the data as those stakeholders with more resources. It was unclear who would be involved in the communication aspect of this project and the group thought that the scientists doing this research should not lead this effort. DNTP should actively engage with the stakeholders that will use the updated database, such as putting out a call for proposals, to ensure this effort is done in a way that will be useful for the end users. He noted that there is interest among the general public for the EO-CRC database, especially among the younger generation. DNTP should also make a point of engaging with the communities that experience health disparities. Dr. Chiu questioned whether mutational signature was a distinguishing aspect of the etiology of these tumors given that many pathways lead to tumor development.

5.2.2. Second Question

The disease-focused approach of the Health Effects Innovation Programs is novel in toxicology and hazard assessment. What unique challenges are we likely to encounter in taking that approach for carcinogenesis? What near and mid-term outputs will build confidence in using that approach?

Dr. Berube, the first discussant, identified himself as a science communication professional. He cited the literature on habit formation, which is a powerful driver in why experts do not take advantage of new protocols, even when they are superior to those they have used in the past. This is called the status quo bias or availability bias. Even experts have confirmation bias and tend to read the literature from their own personal perspective. That is a challenge for the Carci HEI program. He also described trans-cultural issues as obstacles to the success of the approach. He discussed the phenomenon of "promotional science," which is seen as an extension of the public relations and advertising world. This is historically problematic since scientists are uncomfortable with promoting their work. He would like to see stages of ongoing evaluation built into the model. He noted that communications need to take place where professionals get together, with a strong spokesperson who can speak the language. He described work he had been conducting on social media fatigue and cautioned that it will be a challenge if social media is chosen as a platform for getting the message out.

Dr. David Michaels, the second discussant, said that the key is the introduction of actionable information. The field is comfortable interpreting the results of long-term animal studies, even in the absence of epidemiology studies. It is still challenging to interpret the meaning of long-term studies, and as the field progresses and moves to different intermediate points in the carcinogenesis process, there will be increasing challenges and controversies. Translation to the public is another challenge. As the disease-focused approach is being developed, there should be engagement with different types of consumers of the information. He cited the example of shampoo as involving a variety of stakeholders – manufacturers of the shampoo, salon workers, and people who use the shampoo – who would need actionable information to know about any hazards associated with their use.

Dr. Matthew Martin, the third BSC discussant, approved of the disease-focused approach, but expressed concern about a core lack of translation. DNTP might be especially capable of using some of novel tools such as spatial transcriptomics, which has seen much development and would seem to be pertinent in cancer research. EO-CRC is a good choice of concept for building confidence in the *in vitro* models of cancer development. The longer-term challenge is where to go next. He agreed with the EWG comment on the importance of chronic inflammation and immunology in cancer research.

Dr. Singla said that one of the challenges of the disease-focused approach is starting with what is known about the disease, and what is known has been heavily genetically focused. It is unclear what proportion of substances contributing to carcinogenesis can be identified through that approach. Thus, it is important to integrate approaches looking at the key characteristics of carcinogens sooner rather than later to expand the scope of inquiry so that it encompasses a broader variety of substances and the mechanisms by which they may contribute to carcinogenesis. Identifying such substances would be an output that would build confidence in the disease-focused approach.

Dr. Wang said that the points raised by the discussants were well taken and had been discussed internally. She noted that the presentation did not cover all of the projects underway and provided assurance that the team is not solely focusing on mutations or genotoxicities. She pointed out that the National Institute of Environmental Health Sciences (NIEHS) had recently established an Environmental Health Disparity faculty to start tackling some of the issues cited by the discussants. She agreed with Dr. Berube that it is important to go to the stakeholders since that is where they are the most comfortable. She described an ongoing study on chemicals in hair dye and indicated this was similar to Dr. Michael's example of shampoo.

Dr. Germolec noted that there were several DNTP projects underway in the areas of immunology, chronic immune suppression, and chronic inflammation.

5.2.3. Third Question

A key theme of the NIEHS Strategic Plan is "Data to Knowledge to Action." An essential part of the Carci HEI program includes developing a panel of alternative assays (to be used alone, prior to short-term or subchronic in vivo studies, or to complement in vivo assays) to provide mechanistic information and predict in vivo carcinogenicity. What factors should be considered in selecting existing or novel in vitro assays, models, technologies, and platforms? With whom should we consult towards identifying a path forward? What innovative ways do you suggest we use to gain input on our research strategy?

Dr. Felter, the first discussant, encouraged DNTP to elaborate on what is meant by developing a panel of assays to predict *in vivo* carcinogenicity. In the more traditional approach, the focus was testing up to the maximum tolerated dose, regardless of what it was in relation to potential human exposures, with human relevance considered as a separate step after completion of the bioassay. DNTP has made huge progress in shifting the question toward human relevance. She encouraged DNTP to think about the goal—whether it is to predict carcinogenicity under any condition, or to predict conditions that could lead to an increased human-relevant cancer risk. She referred to IVIVE and the kinetically derived maximum dose, and encouraged DNTP to think of building those into assay development upfront, regardless of the actual endpoint. She

discussed which endpoints DNTP should focus on, such as the true role of genotoxicity and mutagenicity for human cancer risk. DNTP should not exclude further investigation of genotoxicity and mutagenicity as new assays are being considered.

Dr. Ryan, the second discussant, noted that from a preclinical perspective, DNTP is well-positioned in terms of collaborations to develop and evaluate alternative assays. She alluded to a reference that showed 18 positive compounds for EO-CRC in rats and mice and encouraged DNTP to think about how those compounds would be prioritized. She cautioned about external collaborations, to not have them be too dilutive from the primary goal, which is bridging tissue and disease. The Carci HEI program concept document had referenced "presumptive environmental exposures" and "populations at risk," but did not specify evidence or details. Dr. Ryan noted that the concept also discussed relying on the literature for human mutational analysis of EO-CRC, but she cautioned that as much as 50% of those tumors have a hereditary component. In addition, EO-CRC has an unclear etiology, with several co-morbidities and indirect effects such as microbiome alterations, so it will be challenging to tease out the chemical exposures that may contribute to the pathogenesis. She suggested exploring linking the EO-CRC mutations with data around environmental exposures. There may be opportunities for collaborations with investigators with well-described EO-CRC cases, where the hereditary component could be eliminated.

Dr. Eaton added that it would be important to include consideration of the role of diet in EO-CRC.

Dr. Eric Blomme, the third discussant, suggested a focus on robust models that target specific mechanistic endpoints. He discussed tissue differences in terms of the types of mutations. Likewise, *in vitro* models will see differences depending on the cell of origin. A challenge is that the majority of the *in vitro* models developed are not that useful and therefore, DNTP should take the time to validate and characterize these models. Metabolic activation should also be kept in mind. He approved of a well-curated database of the key genotoxic agents and non-genotoxic carcinogens, which would be a great resource for the community, and hoped DNTP would lead in setting standards for *in vitro* model validation, allowing correlation, which is currently lacking. He also hoped that DNTP would take the lead on integrating the microbiome as part of the evaluation of models. In terms of communication, publication in the top journals is the best way to generate feedback and get ideas, as well as presentations in the top meetings. He cited the example of the recent Nature Genetics paper. He recommended that DNTP engage with as many collaborative partners as possible.

Dr. Eaton noted that he had not heard reference thus far to the role of the colonic microbiome in its role in forming mutagenic substances in the colon or modifying environmental factors to become mutagenic substances.

Dr. Pandiri clarified that consideration of the role of mutational signatures does not focus on mutagenic aspects. Also, the project is looking specifically at sporadic colonic cancer and not any of the linked syndromes. Regarding the microbiome, the team is looking at the literature, including recent publication suggesting that antibiotic use may contribute to later development of colon cancer due to alterations in the microbiome. DNTP and the National Center for Toxicological Research are actively working on microbiome investigations.

6. Combined Exposures and Mixtures Program

Dr. Cynthia Rider briefed the board on the Combined Exposures and Mixtures (CEM) Program. She introduced the Program Management Team, which consisted of Drs. Kembra Howdeshell, Jui-Hua Hsieh, Cynthia Rider, Suramya Waidyanatha, and Nigel Walker, and described the program's Problem Statement:

- Characterizing exposure to mixtures, evaluating their toxicity and hazard, and assessing associated risk presents challenges.
- There is inconsistent use of available mixture methods and uncertainties in their application.
- Lack of harmonized terminology and methods comparisons complicate information synthesis and impede the use of mixtures data in decision-making.

Dr. Rider provided definitions of terminology in the field and a historical perspective of achievements and progress made by NIEHS/NTP regarding mixtures and combined exposures. She outlined three program objectives:

- Objective 1: Developing and applying a disease-centered systems biology approach for prioritizing mixtures for toxicological and hazard characterization to inform cumulative risk evaluation.
- Objective 2: Developing and applying methods for complex mixture testing and data interpretation to inform risk assessment of whole mixtures.
- Objective 3: Applying component-based approaches by experimentally evaluating defined mixtures and using predictive modeling approaches (e.g., dose addition, response addition) and comparing the results with alternative whole mixture evaluation.

Under Objective 1, the CEM program is developing and applying a new method for determining which chemicals to include in mixtures risk assessments. Current methods of grouping chemicals based on similar mechanisms of action or co-occurrence are not necessarily the most protective or the most scientifically sound. A disease-centered approach focusing on diseases (e.g., cancer, cardiovascular disease) that are priority areas of interest for DNTP will be used to test the hypothesis that chemicals that target disparate signaling pathways contribute cumulatively to disease development, and their joint action can be estimated using mixture modeling approaches.

Objective 2 focuses on applying targeted and non-targeted chemical analyses, *in vivo* bioassays, and literature review methods for complex mixture testing and data interpretation to inform risk assessment. It includes developing methods for complex mixture evaluation including sufficient similarity, polypharmacokinetics, and bioassay-guided fractionation to identify toxic constituents. In addition, this objective aims to provide DNTP research support for the Botanical Safety Consortium—a public-private partnership aimed at developing a toolbox of *in vitro* assays for identifying hazards associated with botanical ingredients.

Objective 3 seeks to strengthen component-based approaches, the current default approach for mixtures risk assessment, which incorporate dose-response data from individual chemicals to predict mixture effects. Dr. Rider described ongoing studies with polycyclic aromatic compounds that are aimed at evaluating the assumptions inherent in component-based approaches.

Dr. Rider outlined the many stakeholders involved with the Converging on Cancer initiative, the DNTP Botanicals Program, and the DNTP Polycyclic Aromatic Compound Mixtures Assessment Program, and referred to the anticipated milestones associated with the programs.

There were no clarifying questions from the BSC members.

6.1. Public Comments

Dr. Eaton noted that EWG submitted written comments on this topic that were distributed to board members.

Dr. David Andrews, a senior scientist from EWG presented recommendations regarding the CEM Program. EWG supports DNTP efforts to take on the issue of mixtures. They recommend that DNTP focus on the real-life exposures that come from mixtures, as an effort to prioritize and target chemical combinations for in-depth analysis. The availability of published information and datasets that already exist is a way to prioritize mixtures for an informed cumulative risk evaluation. He cited several examples of existing information to draw upon, such as purchasing data for consumer products, pesticides frequently detected in fruits and vegetables, and chemicals identified by biomonitoring in people. Using targeted approaches and biomonitoring datasets will help to meet the critical need for progress in the area of mixtures risk assessment. It is important that there are concurrent studies relating to the key characteristics of carcinogens and the impact of combined exposures. There is the underlying concern that different chemical combinations can lead to increased risk of carcinogenesis.

6.2. BSC Discussion

Board members were asked to consider three questions.

6.2.1. First Question

Consider the Problem Statement, Objectives, and Value Proposition in the Program Concept document.

- ***Group #1: Share your insights regarding whether there is clear alignment among the three. For example, do the Objectives align with the Problem Statement? Does the Value Proposition match what is being stated in the Problem Statement?***
- ***Group #2: Share your insights on the strategic fit of the overall program with DNTP's mission, goals, and capabilities.***
- ***Group #3: Share your insights on whether there is sufficient focus to deliver the intended value to stakeholders.***

Dr. Eaton asked Dr. Mader to facilitate the exercise for board members. She read the question and said the BSC members would break into three work groups to discuss the three topics listed above. Following the groups' deliberations, Dr. Mader called upon the group leaders to report on their results.

Dr. Martin provided input from Group #1's discussion (see Attachment D for slides presented). The group agreed that this was a very complicated issue and appreciated that DNTP focused on defining the terminology used in this field. The approaches used (e.g., traditional, practical applications, conceptual development) aligned well with the objectives and problem statement.

The group thought DNTP focused too heavily on pharmacodynamics and not enough on pharmacokinetics for these complex mixtures. More clarity was needed on how the read-across approaches would be applied to mixture risk assessments. DNTP should elaborate on the process for developing the disease-based project for mixtures; this would provide clarity on the relationship between the Carci HEI and CEM programs. The group was surprised that contaminants in water were not addressed as an example of a real-world mixture. Further explanation is needed on how the interaction between non-chemical stressors and mixtures exposure would be incorporated into the research program.

Dr. Pamela Lein added that it would be important to communicate the complexity of mixtures to stakeholders and the lay public. Additionally, she stressed why it is important to start with simplified, well-defined mixtures to build understanding about how interactions between mechanistically diverse classes of chemicals lead to disease, and how non-chemical stressors impact the effects of the chemicals. Dr. Chiu noted that the decision context for researching botanicals was clearly stated; however, DNTP should emphasize how the results from the polycyclic aromatic hydrocarbons (PAHs) research could impact decision making.

Dr. Singla provided input from Group #2's discussion (see Attachment E for slide presented). She noted that DNTP's mission is focused on improving public health by performing research that aligns with real-world public health needs and translating those findings to inform better public health outcomes. The group agreed that this area of research was essential for public health need and potential impact. There was, however, a lack of cohesion around specific areas of the program and the impact on public health. For example, what is the disease burden of botanical supplements, who is most affected by botanicals, and is more research on wood burning stoves the key to action for public health outcomes? The group thought DNTP's efforts were spread too thin and there was a lack of cohesion among the projects that were highlighted from this program. More clarity is needed on the priorities. The group agreed that it was important to continue research on PAHs. DNTP should clarify the problem statement to focus on the real-world public health problem, the ultimate outcome the research will inform, and the populations and diseases the program will impact. It should be clear that risk assessment and methods are not the ultimate goal, rather those are tools to improve public health outcomes and decisions.

Dr. Blomme provided input from Group #3's discussion (see Attachment F for slide presented). The group agreed this was a complex problem to tackle. They were worried that this research may not be realistic but understood that what was presented was only part of the picture. The current component-based approach is not ideal, and the group agreed that DNTP should move towards using a more mechanistic evaluation. In the pharmaceutical environment, there are a series of robust, well-characterized models and assays to study compounds prior to moving to phase 1 trials in humans. Even in this well-controlled environment with just one chemical, the researchers may not get it right. Thus, it is important to keep realistic expectations when investigating multiple compounds together in an environment that is not as well-controlled. This is a very difficult and complex problem to undertake. More clarity is needed for Objective 1 and how the disease-centered approach will play a part in CEM.

Dr. Rider noted that part of the confusion was around the examples that had been provided, but the actual aim of the program is to identify underlying principles of mixtures' hazards. The botanicals were used as an example of a complex mixture to build the tools to apply to other

complex mixtures. For example, the approaches used with polycyclic aromatic compounds could be applied to per- and polyfluoroalkyl substances (PFAS) as a group. The disease-centered approach is trying to understand what should be included in a cumulative risk assessment and where chemicals should be prioritized because they are likely to act additively towards disease development. The exact chemicals that were discussed are not as important as trying to uncover the underlying principles in the approach. The objectives address different areas of mixtures research.

Dr. Kembra Howdeshell added that the program is diverse because DNTP has a very diverse portfolio. She noted that the group started with the existing mixtures portfolio and organized the ongoing projects into the three areas described in the objectives. The mixtures strategy helps hone the projects so that the program is addressing important methodologies that will be applied to future projects.

Dr. Eaton noted that the board appreciated the magnitude of the challenge in addressing mixtures.

6.2.2. Second Question

Now refer to the sections on Stakeholders and Milestones in the Program Concept document. With whom else might we partner to ensure program success? Discuss whether the proposed activities and outputs demonstrate that the program is achieving the objectives. What other metrics might better define the value we envision?

Dr. Mark Russi, the first discussant, noted that with some of the well-established carcinogens, the initial signal was from occupational epidemiology and working populations. In terms of collaboration, he wanted to be certain that there is adequate collaboration with the National Institute for Occupation Safety and Health and with centers looking at large occupational cohorts. These collaborations will help ensure adequate monitoring of the emergence of new signals. It is important that the project be prioritized adequately, as there are societal prioritizations based on burden of disease.

Dr. Singla, the second discussant, agreed that it would be important to think about priorities in partnerships and collaborations in terms of thinking about populations experiencing real-world multiple chemical exposures disproportionately. She agreed with the concept of using case studies to characterize particular classes of chemical compounds but indicated there may be a better return on investment if the case studies were more aligned with some of the public health priorities so that DNTP could develop principles and share knowledge concurrently.

Dr. Lein, the third discussant, said that mixtures is a complex issue and that DNTP is the right organization to take this on. She recommended partnering with the Environmental Health Sciences Centers program funded by NIEHS, as many of the Centers have ongoing research in mixtures and toxicology. For example, she mentioned one NIEHS-funded center that is studying wildfires and has data on health outcomes, populations exposed to wildfires, and the compositions of the ash and smoke. This would align with the PAHs research. With new technologies being developed by some of the centers looking at mixtures, there are several opportunities to collaborate to encourage the systems biology approach.

In response to Dr. Singla's comment, Dr. Rider noted that 18% of the population takes botanicals, often at very high doses. In addition to being a public health concern, botanicals have provided DNTP the opportunity to develop methods to evaluate other complex mixtures.

Dr. Lein agreed with Dr. Rider's comments regarding botanicals and noted that there is no research on how botanicals influence the response to other toxic agents.

Dr. Singla appreciated the additional information on botanicals. She clarified that she was not stating that botanicals are not an important area of research for public health but questioned whether they are the most important priority. Disproportionately impacted populations in terms of multiple chemical exposures should be a priority factor.

Dr. Eaton noted that botanicals are a topic of wide-ranging research among a variety of stakeholders, so it is not something DNTP is taking on in isolation.

6.2.3. Third Question

A key theme of the NIEHS Strategic Plan is "Data to Knowledge to Action." There are both unique opportunities and challenges in the use of traditional and novel testing and assessment approaches. How should we focus our efforts to maximize the impact of DNTP combined exposures and mixtures research in the field? How will we know when we have achieved the right balance in research aimed at refining established methods, such as component-based assessments, with exploration of novel approaches that have not yet been widely adopted by the public health/risk assessment communities, such as those focused on whole mixtures and specific diseases, exposure scenarios or populations?

Dr. Chiu, the first discussant, liked the suggestion that a potential application for PAHs would be in wildfires and discussed the importance of finding a real-world exposure partner to demonstrate the application for Data to Knowledge to Action. Getting to action should be part of the conceptual development of the disease-focused approach. Overall, the balance was good in terms of the various approaches.

Dr. Gray, the second discussant, remarked that the complexity of the work is a barrier to public health action in terms of the ability of a public agency conducting research to help inform actions in the community – this challenges the team to be very intentional from the outset. She would like to see a more explicit connection to water, for example, particularly regarding the presence of PFAS, a major emerging contaminant in water that is of concern in communities across the county. She reiterated the importance of the interplay with non-chemical stressors, especially in communities with high racial and ethnic diversity and lower income, who are bearing an undue burden of disease due to environmental contamination. Exposure to multiple contaminants is often a concern in those communities. She was excited about the work because it directly addresses questions she hears often in the communities she works with and encouraged the team to be as transparent as possible about why they are making the choices they make.

Dr. Rider thanked the board members for their valuable feedback. Vulnerable populations would be kept in mind while the team is designing studies, as well as the importance of chemical selection.

Dr. Howdeshell noted that there are some efforts within DNTP to better understand exposures and the relevance of the doses being used, as well as to better understand how projects impact

communities facing challenges such as health disparities. She asked Dr. Gray for suggestions on how DNTP can better reach out to communities with regard to mixtures projects. Dr. Gray recommended that DNTP reach out to the NIEHS extramural community and stakeholder partners as good networks to tap into for community engagement. Dr. Eaton, a former director of one of these NIEHS centers, agreed that this was an excellent avenue to reach the public.

Dr. Martin said there is a need to develop standards for mixture metadata to better describe them for inclusion in databases. Dr. Rider said the team has recognized the need for a dedicated mixtures database.

Dr. Eaton discussed the challenge of dose as it applies to multiple compounds, which adds another layer of complexity when trying to figure out real-world interactions.

7. Adjournment

Dr. Eaton turned to Dr. Woychik and Dr. Berridge for their closing comments.

Dr. Woychik thanked everyone for the insightful comments during the meeting, and thanked Dr. Wolfe and the DNTP staff for their hard work in organizing the meeting. Dr. Berridge added his thanks to everyone for their contributions. Dr. Woychik thanked Dr. Eaton for chairing the meeting.

Dr. Scruggs thanked the board members for their active participation in the meeting.

Dr. Eaton adjourned the meeting at 4:55 pm.

8. Approval of the Summary Minutes by the NTP BSC Chair

These summary minutes have been read and approved by the chair of the February 2, 2021 NTP Board of Scientific Counselors.

David Eaton, PhD, University of Washington

NTP BSC Chair

Date: May 6, 2021

9. Attachments

Attachment A

Question 1: Carcinogenicity Health Effects Innovation Program

Group #1: Share your insights regarding whether there is clear alignment among the three. For example, do the Objectives align with the Problem Statement? Does the Value Proposition match what is being stated in the Problem Statement?

- Project on early onset colorectal cancer (CRC) is of high priority. It was not clear which partnerships were already existing in this work. A lot of groups that we did not have time to talk about. Ensure that NTP is fully connected with those doing research, such as in dietary restrictions.
- Great example of clear alignment of how NTP can build science that addresses science impacting human health. Not just bench research, but impacting human health.
- Questioning whether in vivo approach link to or inform the in vitro approach and whether those approaches are informative of the human condition
- Good approach, clear difference between minority groups.
    - Two weaknesses: Microbiome, rat models
    - Early onset CRC is distinguishing itself
    - Rat models: APC mutation, linking early onset CRC to
    - Parallelogram approach, building linkages. Molecular signatures from CRC patients is a critical part of how NTP is planning how to ensure their research is relevant to what is seen in human patients
    - Organizing an approach

- Problem statement was right on in terms of identifying the problem and the focus
  - Calling out disproportionately impacted groups
  - Disconnects: the objectives and approaches therein, how impactful those particular objectives relate to the overall problem
  - Unclear how approaches/objectives are incorporating the exposures, particular types of cancer in disproportionately affected groups
  - Question: How can this research be relevant to the real-world conditions of disproportionately affected groups?
    - Very likely that these groups have particular kinds of exposures
  - Doing research in partnership with organizations that have specific knowledge on those groups to ensure that the research reflects the factors that are important to the groups
    - EPA Environmental Justice Office

- Like to know more about purpose-built platform
  - Variables that go into platform
  - Get information to the public as quickly as possible and consider prioritizing in a timetable structure or find a way to integrate views of President Biden

Attachment B

Question 1: Carcinogenicity Health Effects Innovation Program

Group #2: Share your insights on the strategic fit of the overall program with DNTP's mission, goals, and capabilities.

- Thinks this fits, in preclinical space with collaborations to develop new capabilities. Bridging to human disease and collaboration with public stakeholder could strengthen this. (Anne)
- NTP is fundamental to identification of new carcinogens, how do we bridge the translation of this for stakeholders. Fits well into overall objectives. (David M)
- Overall, this fits nicely into overall NTP mission. Areas of expansion: how to interpret to human health and how training will fit into overall mission. Gene by environment interactions. (Pam)
- Missions and goals align with partnerships (internationally) and sequencing tech for mutational spectra. (David E)
- Mixtures are complex in how they drive cancer, is there active consideration of this for application to the cancer program? How do programs mixtures and cancer programs interact? (Pam)
- As the scientific community thinks of NTP through history, how are programs fitting into standard 2-year rodent bioassay? More on what NTPs role will be in these assays as a goal of NTP is to replace these? How does this integrate with NTPs plan to replace these assays? (David E)
    - Plan to address this going forward. Opportunity to build in mechanistic knowledge/understanding. (David E/Anne)

Attachment C

Question 1: Carcinogenicity Health Effects Innovation Program

Group #3: Share your insights on whether there is sufficient focus to deliver the intended value to stakeholders.

- Do not have enough information to answer these questions because we are unsure how you define “intended value”?
- Do stakeholders with less resources have the same opportunity to access the same data as those with more resources?
  - Who are the priority stakeholders for this team?
- Who is involved to help deliver the message to the stakeholders?
- Regarding the databases, have you engaged the stakeholders that will use this information to make sure the data is curated in the best way possible?
  - Potentially put out a call for proposals on how to use these tools (active engagement)
- The colorectal database is useful for stakeholders in the general public
- Etiology of cancer – there is a large group of stakeholders in the research community that will find this information useful
- How are you responding to the disparity between those develop cancer and how are you engaging these stakeholders?
- Communication aspect seems critical for all programs not just Carci

Attachment D

Question 1: Combined Exposures and Mixtures Program

Group #1: Share your insights regarding whether there is clear alignment among the three. For example, do the Objectives align with the Problem Statement? Does the Value Proposition match what is being stated in the Problem Statement?

- Overall, yes they align.
  - This is very complicated issue and NTP's approach is as good as any others.
  - There was a lot of emphasis on interactions at PD level but not so much on the PK level.
  - Some confusion around read-across approaches.
    - Would appreciate more clarity on how the read-across approaches can be applied to a class of compounds (or even in a mixture itself).
  - Getting clear communication on what methods should be applied is useful.
  - #3 is a fairly traditional approach (PACMAP).
  - #2 has very direct and practical applications.
    - Could approach on #2 be applied to #3?
  - #1 is still in conceptual development.
  - What is the process for developing the disease-based project?

- What is relationship between Carci and CEM?
  - A great opportunity for program staff to communicate.
- Surprised that contaminants in water was not addressed.
- You have mechanistic classes in real-life chemicals.
- Understand that it is a complex issue!
- Whole interaction of non-chemical stressors and their impact.

Attachment E

Question 1: Combined Exposures and Mixtures Program

Group #2: Share your insights on the strategic fit of the overall program with DNTP's mission, goals, and capabilities.

- Overall area of research is a good fit for public health need and impact. Disconnect around specific areas of investment and impact for public health needs. Where is public need for botanicals and wood burning stoves research? (Veena)

- Are NTPs efforts becoming diffuse? Taken on many activities and put into buckets because they seem to match. Sounds like many projects are ongoing, but difficult to determine NTP scope/capacity to take on this work. (Anne)

- Aligns with previous comments. Priorities typically come from epidemiological signals, often from distinct exposures. Glad to see PAHs cited, there are highly exposed working populations and is a rich area. Pay attention to epidemiological signals and how much burden a disease may be to society. (Mark)

- Something was missing from problem statement, clarifying the statement may help focus statement. Problem is mixtures are contributing to disease, assessments accounting for mixtures are not accounting for this. Get to public health problem and outcome with mixtures – call out specific populations (international? Within US?). What population is of interest here. (Veena)

    - Agree (Anne)

- Bring together the impact/outcome to the narrative of the program. In reading the document, it seemed to be risk assessment in the end; this is a tool to achieve better public health outcomes but is not the end goal. How do you get accurate assessment of risk? Need to have the ultimate outcome in mind. (Veena)

Attachment F

Question 1: Combined Exposures and Mixtures Program

Group #3: Share your insights on whether there is sufficient focus to deliver the intended value to stakeholders.

- Worried that approaches focused on adding individual components together may yield an unrealistic estimate of the true risk
- How are we defining mixtures? What humans are exposed to in the environment is very complex
- Looking at individual chemicals or multiple mixtures that can be additive
- Component based approach may not be ideal and needs refinement
- Long history of drug-drug interactions, encourage using a mechanistic approach to studying mixtures
  - Picking the mechanism may be tough, encourage looking how mixtures modify a known MOA
- How much can you generalize from these individual examples?
  - Maybe make a system of these interactions
- What is feasible when studying these mixtures, this is very ambitious
- How is disease-focused playing a part in this group? (Objective 1)
  - Are you starting with cancer/cardiovascular disease in humans or were those just examples?
- Will need to look at multiple possible outcomes in mixtures to make sure you don’t miss anything