NTP Research Concept:
Polycyclic Aromatic Hydrocarbons (PAHs)

Project Leader

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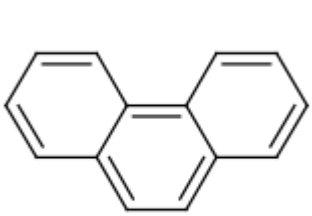
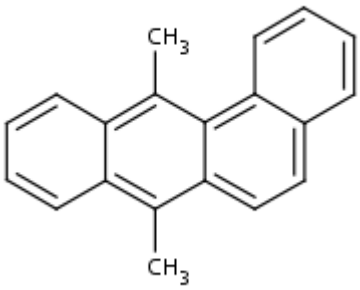
Background and Rationale

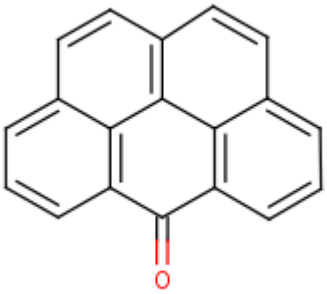
Figure 1. Select examples of polycyclic aromatic hydrocarbon (PAH) structures:



Unsubstituted PAH
Phenanthrene
CAS No. 85-01-8



Alkylated PAH
7,12-Dimethylbenzanthracene
CAS No. 57-97-6



Oxygenated PAH
6H-Benzo(cd)pyren-6-one
CAS No. 3074-00-8

Chemical identification and toxicity

Polycyclic aromatic hydrocarbons (PAHs) are composed of two or more fused rings made up of carbon and hydrogen atoms (see IPCS 1998 in supporting documents). They have at least one aromatic ring and can contain 5-membered, non-aromatic rings. The PAH chemical class includes unsubstituted and alkylated compounds and belongs to a broader polycyclic aromatic compound (PAC) class that also includes N-, S-, and O-substituted PAHs as well as heterocyclic compounds containing N, S, or O atoms within the ring structure. For the purposes of this document, PAHs and related compounds are referred to generally as PAHs or by a description of the defining structural modification (e.g., oxygenated PAHs, N-heterocyclics).

PAHs are ubiquitous environmental contaminants occurring naturally in crude oil or created and released into the environment through natural events (e.g., volcanic eruptions, forest fires) and anthropogenic activities (e.g., burning of fossil fuels). The processes by which PAHs are created and move through the environment result in complex and dynamic mixtures. Human exposure to mixtures of PAHs can occur through consumption of PAH-containing foods (e.g., contaminated seafood, char-grilled meat), non-dietary ingestion (e.g., house dust), inhalation of polluted air (e.g., cigarette smoke, diesel exhaust), or dermal contact in an occupational setting (e.g., road paving, roofing). In order to estimate the risk from exposure to real world PAH mixtures, it is necessary to understand the cumulative effects associated with this class of

compounds. However, there are multiple factors that contribute to the complexity of this challenge:

- The class is large (>1500 identified PAHs) with compounds having diverse structural features (ring configurations and substitutions).
- Individual PAHs target multiple cells and tissues (e.g., genotoxicity, immunotoxicity) and act via disparate mechanisms of action.
- PAHs exist in complex and dynamic mixtures that often contain other known environmental toxicants (e.g., dioxin-like compounds, heavy metals).
- Exposure to PAHs can occur via multiple routes (i.e., dermal, oral, and inhalation) and in both gaseous and particulate phases.

PAHs display a wide range of toxicities and a complicated array of mechanisms of action. The International Agency for Research on Cancer (IARC) has classified benzo(a)pyrene (BaP) as well as multiple occupational exposures to PAH mixtures (e.g., coal gasification or coke production) as Group 1 human carcinogens (see IARC 2010 in supporting documents). Additionally, multiple individual PAHs are classified as Group 2A (probably carcinogenic to humans) or 2B (possibly carcinogenic to humans) carcinogens (IARC 2010). Lungs, bladder, and skin are common sites for tumors associated with PAH exposure [1]. The mechanisms of carcinogenicity associated with PAHs encompass both initiation and promotion phases of the carcinogenic process. PAHs can be metabolized by various phase I enzymes to reactive metabolites that are capable of damaging DNA. Additionally, some PAHs bind to the aryl hydrocarbon receptor, which likely plays a role in mediating both genotoxic (through inducing enzymes that form reactive PAH metabolites) and non-genotoxic (through dysregulation of cell cycle control [2]) carcinogenesis pathways. Other mechanisms involved in PAH-mediated potentiation of carcinogenesis include interference with gap junction intracellular communication [3] and perturbation of cell signaling [4].

Although the majority of PAH research has focused on characterizing the carcinogenic effects and mechanisms of carcinogenicity, there is substantial evidence that PAHs also elicit multiple non-cancer toxicities. There is a significant body of work demonstrating the immunotoxicity of select unsubstituted PAHs, characterized by suppression of humoral immunity [5]. Reproductive toxicity has been demonstrated in animal models with BaP and in humans exposed to complex mixtures of PAHs. BaP has been found to elicit ovarian follicle loss in female mice [6, 7], while decreased sperm quality has been observed in both animal models and human populations exposed to PAH-containing mixtures [8-10]. Developmental toxicity characterized by a reduction in fetal birth weight and length and impaired intrauterine growth has been observed in PAH-exposed populations [11, 12]. Also, neurotoxicity as measured by increased anxiety [13] and aggression [14] in animal models and increased behavioral problems in humans [15] has been associated with PAH exposure.

<u>Knowledge gaps</u>

Despite the significant challenges outlined above, risk assessments for PAH-containing mixtures are regularly conducted to estimate the potential health effects associated with exposure to PAHs at contaminated waste sites, in air pollution, or in determining

acceptable levels of contamination of certain foodstuffs such as seafood. The basic strategies available for estimating the combined effects of mixtures can be classified into two broad categories: whole mixture and component-based. Whole mixture approaches involve characterizing the toxicity of a complex mixture of interest or comparing an unknown mixture to a well-characterized reference mixture that is deemed to be sufficiently similar based on chemical and/or biological criteria. In contrast, component-based approaches utilize individual constituent dose-response data to predict the toxicity of the mixture. There is on-going debate regarding whether a whole mixture or component-based approach is more appropriate to estimate the effects of PAH mixtures. Although there is a lack of adequate information on the overwhelming majority of individual PAHs, the component-based approach is currently recommended by the US Environmental Protection Agency (EPA) as the default, due to the even greater dearth of data on whole mixtures. A component-based approach was also used by the FDA to assess the safety of seafood in PAH-contaminated Gulf of Mexico waters following the 2010 Deepwater Horizon Oil Spill (see FDA 2010 in supporting documents).

Individual PAHs, as well as the class as a whole, have been nominated on multiple occasions to the NTP for toxicological evaluation with different data gaps specified by the various nominating parties. These nominations generally cite the lack of adequate toxicity data for assessing public health risk associated with exposure to complex PAH mixtures. ***The knowledge gaps associated with estimating the health effects of PAHs fall into the following interdependent categories:***

1. **<u>Exposure characterization</u>**
   There are approximately 16 commonly monitored PAHs representing the class of 1500 plus compounds. These 16 PAHs are frequently monitored because they are included on the EPA list of "priority pollutants" that are regulated under the Clean Water Act. It is unclear whether these are the most appropriate indicator compounds, or simply the most robustly characterized PAHs. In terms of exposure to complex environmental mixtures, any PAH-containing environmental mixture is unlikely to be fully characterized (i.e., some unidentified fraction will remain) regardless of the effort expended in its chemical analysis.
2. **<u>Hazard characterization</u>**
   The bulk of PAH research has focused on relatively few PAHs. The endpoint most frequently assessed is carcinogenicity. The vast majority of PAHs are uncharacterized, and many of those characterized for their carcinogenic potential have not been evaluated for other toxicity endpoints.
3. **<u>Risk characterization</u>**
   To date, risk assessment efforts have focused almost exclusively on carcinogenicity associated with select commonly monitored, unsubstituted PAHs and have conformed to the relative potency factor (RPF) approach. The RPF approach is a component-based approach that relies on the concept of dose additivity, whereby doses of constituent chemicals are converted to reference chemical equivalents (e.g., BaP equivalents) using a conversion factor that takes into account the different potencies of the individual PAHs (e.g., PAH_i slope ÷ BaP slope; where PAH_i is the individual PAH "*i*") and then added together to get

a total mixture dose. The total dose of calculated BaP equivalents for the mixture is then used to estimate the toxicity of the mixture. This approach requires single chemical dose-response data, which is only available for a limited subset of PAHs. Additionally, the assumptions inherent in application of the RPF approach include: dose additivity of PAHs, a lack of interactions among constituents (both among PAHs and between PAHs and non-PAH constituents), a common mechanism of action, and validity of route-to-route extrapolation. An alternative whole mixture approach has not been a viable option as yet due to a lack of adequate whole mixture data, the absence of accepted methods for determining sufficient similarity, and the fundamental unknown of whether or not PAHs are the fraction driving the observed toxicity of complex mixtures.

The knowledge gaps and challenges outlined above have been recognized by other research groups working to inform the PAH risk assessment process. Health Canada, for example, has a significant PAH research program focused on strengthening the scientific basis for conducting cumulative PAH human health risk assessments. Scientists at Health Canada have tested 9 unsubstituted PAHs: BaP, benzo(b)fluoranthene, benzo(k)fluoranthene, benzo(*ghi*)perylene, chrysene, benz(a)anthracene, dibenz(a,h)anthracene, dibenzo(a,l)pyrene, and indeno(1,2,3-cd)pyrene, as well as several mixtures (e.g., coal tar, organic extracts of contaminated soil, simplified PAH mixtures) for genotoxicity in 28-day oral gavage studies using the MutaMouse model. The PAH research project proposed in this concept builds upon and is complementary to the research conducted by Health Canada.

Clearly, any manageable PAH research strategy will not address all of the knowledge gaps associated with this complex problem. However, the proposed NTP PAH research program is focused on strengthening the scientific basis for risk assessment of PAH-containing mixtures in a logical and targeted manner. Furthermore, international collaboration and cross-disciplinary expertise will be leveraged to build a maximally productive PAH research program. The research will contribute to each of the interdependent categories described above (i.e., exposure, hazard, and risk characterization). This program will incorporate a deliberately iterative and flexible design, which will allow for intermediate evaluation and, as needed, course-corrections.

Key Issues

1. Exposure characterization

Although there is an extensive literature on exposure to PAHs from various sources, the overwhelming majority of exposure characterizations focus on the commonly monitored PAHs. For the purposes of the NTP research program, the major underlying questions regarding exposure to PAHs are: (1) Do the 16 commonly monitored PAHs represent exposure to the entire class? (2) Which PAH mixture profiles are associated with specific sources and exposure situations of concern? (3) Are there human (or wildlife) exposures to individual PAHs that have significant hazard potential? The more detailed analyses of PAH mixtures available in the literature (e.g., [16]) will be used to help inform the selection of individual PAHs for study.

Although it is beyond the scope of this project to characterize the extent and nature of human exposure to PAHs, an effort will be made to utilize targeted selection of individual PAHs (including substituted PAHs) and complex mixtures to inform characterization efforts. For example, a determination of the hazard associated with select PAHs that have not been well-studied will help inform future decisions on whether or not they should be included in exposure characterization efforts. Key issues in exposure characterization of PAHs include:

a. There is a deficiency in the characterization of complex PAH mixtures used in toxicity assessments.
b. The majority of the limited carcinogenicity studies available have been conducted with dermal exposure, when real-world exposures often occur through oral or inhalation routes.
c. Data are needed to link the external exposure (e.g., diet, air) to internal dose.

2. Hazard characterization

The three driving knowledge gaps related to hazard characterization are (1) the limited number of PAHs (mostly unsubstituted) that have been characterized, (2) the lack of characterization of PAH-driven effects on non-carcinogenic endpoints (e.g., immunotoxicity, reproductive and developmental toxicity), and (3) the inadequate understanding of mechanisms and pathways of toxicity thereby restricting the development of predictive models (e.g., quantitative structure activity relationships [QSAR]).

a. <u>Expanding the field of characterized PAHs</u>
    i. It is impractical to comprehensively assess the toxicity/carcinogenicity of the 1500+ identified individual PAHs and the practically infinite number of mixtures containing PAHs using the traditional two-year bioassay. Therefore, shorter term animal studies, alternative animal models, and *in vitro* assays are needed to provide information on a broader subset of individual PAHs and PAH mixtures.
        - 60 individual PAHs are included in the 10K compound library being screened in nuclear receptor and stress response pathway assays that are the current focus of the Tox21 high throughput screening (HTS) efforts. Although no PAH mixtures are being screened currently, Tox21 has the ability to screen mixtures in these assays. To assess the utility of the HTS approach, evaluations of data across the included PAHs and between HTS data and *in vivo* results are required. The goal is to identify biologically meaningful endpoints that can be used in development of a PAH-specific toxicity profile(s).
        - Alternative animal models (e.g., zebrafish developmental model) offer great promise and have already been used to assess multiple PAHs. More research is needed to characterize a broad range of PAHs in alternative animal models.

- Targeted in vivo studies are required to link HTS data and findings from alternative animal model studies of individual PAHs and mixtures to mammalian endpoints that are related to human disease.

ii. The PAH class contains a multitude of constituents with different structural features (e.g., unsubstituted; alkylated; O-, S-, or N-substituted; heterocyclic). The majority of the data are from studies that focused on a subset of unsubstituted PAHs. More work is needed to elucidate the impact of structural components on toxicity.

b. Characterizing the spectrum of toxicities associated with PAH exposure

i. Although the majority of research on PAHs has focused on genotoxicity and carcinogenicity, there are other reported toxicological effects (e.g., immunotoxicity, developmental and reproductive toxicity). Additional work is needed to better characterize the spectrum of toxicities elicited by PAHs.

c. Elucidating mechanisms and pathways of toxicity

i. Despite the large number of studies dedicated to elucidating the mechanisms involved in the carcinogenesis of PAHs, these mechanisms are not yet fully understood. Additionally, there has been little research to define the molecular pathways leading to non-carcinogenic toxicities.

ii. The development and validation of predictive models of PAH toxicity are needed to complement testing efforts and increase coverage of the class.

3. Risk characterization

The research described in this concept is focused on gaining a better understanding of the uncertainties associated with application of the current risk assessment paradigm (i.e., the RPF approach) for PAHs and the alternative whole mixture risk assessment approach.

a. RPF Approach

There are four major issues that account for the bulk of the uncertainty associated with the currently prescribed RPF approach for assessing the combined effects of PAHs:

i. There are limited single-chemical data even for chemicals that have RPFs.

ii. The RPF approach has not been thoroughly evaluated for use with PAHs. In effect, studies have not been conducted to compare the predicted response of a mixture based on calculated RPF values to observed mixture responses for relevant endpoints. RPFs are available exclusively for select unsubstituted, commonly monitored PAHs, although over 1500 PAHs have been identified.

iii. RPFs have been estimated for carcinogenicity, while PAHs target additional toxicity endpoints.

b. Whole mixture approach

There are significant uncertainties related to the execution of a whole mixture risk assessment approach that range from identifying appropriate reference

mixtures to developing methods for determining sufficient chemical and/or toxicological similarity of an unknown mixture to the reference mixture(s). Addressing these fundamental issues is beyond the scope of the proposed research program. The key issues that this project will address include:

i. Characterization of select, complex environmental mixtures with broad exposure potential.
ii. Identification of the proportion of toxicity elicited by whole complex PAH-containing mixtures that is due to the PAH fraction.
iii. Determination of whether or not the toxicity of a complex mixture can be adequately predicted by a subset of individual PAHs contained within the mixture.

Specific aims

Although the key issues described above can be loosely divided into distinct categories (exposure, hazard, and risk characterization), they are clearly interrelated. Specific aims developed to advance the understanding of PAH toxicity will necessarily cut across multiple categories and key issues. Therefore, in briefly describing the specific aims of the proposed program, reference will be made to the various key issues addressed by each proposed specific aim.

1. Assess chemical, toxicokinetic (TK), and absorption, distribution, metabolism, and elimination (ADME) properties of select, individual PAHs and mixtures to gain insight into exposure and dosimetry. Key issues to be addressed include: 1.a., 1.b, 1.c., 2.a.i., 2.a.ii., 3.b.i.
2. Characterize the toxicity of a broad range of individual PAHs, defined PAH mixtures, and complex environmental mixtures containing PAHs using a short-term panel that incorporates in vitro, alternative animal, and in vivo models and captures a diverse array of endpoints/effects. Key issues to be addressed include: 1.b, 2.a.i., 2.a.ii., 2.b.i., 2.c.i., 2.c.ii., 3.a.i., 3.b.i.
3. Compare predicted mixture toxicity results using component-based models that incorporate calculated relative potency factors generated from studies described in Specific Aim 1 or whole mixture approaches (e.g., complex mixture fractionation approaches, models based on sufficient similarity of whole mixtures) to observed toxicity. Key issues to be addressed include: 3.a.ii, 3.a.iii, 3.a.iv, 3.b.ii, 3.b.iii.

Proposed Approach

The proposed approach includes four components: (1) targeted selection of test articles; (2) a flexible, iterative format; (3) incorporation of a broad spectrum of endpoints (including *in vitro*, alternative animal model, and mammalian *in vivo* assays) that will capture a range of relevant toxicities in a short-term testing panel, and (4) cross-disciplinary and institutional collaboration. Details on the components of the proposed approach along with potential challenges are described below.

Selection of test articles

Individual PAHs will be selected for study based on the following considerations:

- Environmental exposure potential.
- Structural diversity and likelihood of demonstrating a diverse array of effects (e.g., representative PAHs that are likely to display different patterns of effect based on structure or evidence of divergent toxicological profiles in the literature).
- Representative, commonly monitored PAHs with cancer RPFs assigned with high confidence (i.e., well-studied compounds) that represent a range of potencies (e.g., BaP, phenanthrene) will be included in order to compare results from the short-term panel to results from a wide range of well-conducted studies (e.g., results from the Health Canada PAH Program and available bioassays).
- Representative PAHs that are not commonly monitored in the U.S. and have potent cancer RPFs that have been assigned with low to medium confidence (i.e., RPFs are based on one or two studies). Candidate PAHs include dibenzo[a,l]pyrene with an RPF indicating a potency 30 times BaP based on two studies and benzo[j]aceanthrylene with a potency 60 times BaP based on one intraperitoneal male mouse study. Evaluation of chemicals such as these will help in characterizing the hazard of PAHs that are potentially highly potent genotoxic agents, but remain uncharacterized with respect to other toxicities.
- Representative alkylated (e.g., 1,4-dimethylphenanthrene, 5,9-dimethylchrysene) and oxygenated PAHs (e.g., 9,10-phenanthrenequinone, 6*H*-benzo[*c,d*]pyrene-6-one) will be included based on indications of potential activity from the literature and/or structural features, as well as exposure potential.

Both designed mixtures and complex environmental mixtures will be included in the testing program

- Designed mixtures will be created based on individual chemical data in order to assess the RPF approach.
- Select, complex mixtures will be included in the testing program based on consideration of exposure potential, impact to the overall research program, and public health concern. Selection of complex mixtures will involve collaboration with regulatory agencies to identify the highest priority mixtures and extramural partners specializing in exposure science. The types of complex mixtures that will be considered include, but are not limited to:
    - Extracts collected from passive sampling devices in oil-contaminated waters. These samples could provide an accessible substitute for invertebrate PAH content (e.g., shellfish concentrations).
    - Coal tar extracts tested in the Health Canada PAH program. Use of identical complex mixture samples will allow for cross-laboratory comparison.
    - PAH-containing standard reference materials (e.g., NIST mussel tissue).

Challenges associated with test article selection

Despite efforts to maximize the number of PAH-related test articles that can be included in the program, the total number of individual PAHs or PAH mixtures tested will represent only a very small fraction of existing PAHs or PAH combinations. Therefore, selection of PAHs representing a broad range of chemical properties and biological effects will be important for understanding the scope of the problem. Another challenge

is the difficulty and cost associated with acquiring highly pure individual PAHs that are not readily available and may require synthesis. There are multiple challenges associated with complex environmental samples including acquiring consistent sample in sufficient quantities for *in vivo* assays and conducting in-depth chemical analysis.

Short-term testing panel

The short-term panel will represent the breadth of toxicities that have been associated with PAHs, while maintaining a compact testing paradigm to maximize the number of test articles that can be evaluated. The *in vivo* battery will be designed to build upon the PAH research program at Health Canada focused on genotoxicity, while adding endpoints to characterize additional toxicities (e.g., immunotoxicity, developmental toxicity). An example of potential endpoints to be used in the short-term battery, along with the rationale for inclusion of endpoints, is provided below for illustration.

1. Subacute exposure in male and female rodents (i.e., B6C3F1/N mice and/or Harlan Sprague Dawley rats). Oral gavage is considered to be among the human-relevant routes of exposure [17] and is likely to be the main exposure route, while targeted use of inhalation and dermal exposures will allow for cross-route comparison. The following studies will be conducted for each test article:
    - ADME/TK studies will contribute data to decrease the uncertainty in route-to-route extrapolation.
    - Immunotoxicity cohort – Antigen-specific antibody forming cell assay with sheep red blood cell (sRBC) as antigen. This assay was found to be sensitive for detecting immunotoxic effects of a representative PAH [18] and has been used widely to compare across PAHs [5].
    - General toxicity and genotoxicity cohort
        - Gene expression: liver, lung, and testes.[1] Evaluation of gene expression in multiple tissues will allow for pattern recognition across biological space and could provide information on key pathways affected by PAHs.
        - Pathology: liver, lung, and testes. Observed pathology will provide an anchor for gene expression findings.
        - Hematology: plasma and bone marrow. Changes in hematological parameters will provide information on immunotoxicity.
        - Pig-a gene mutations in reticulocytes
        - Micronuclei in red blood cells

        } Both of these parameters were found to be useful indicators of genotoxicity in the Health Canada PAH program
2. In vitro
    - AhR transactivation assay (e.g., chemically activated luciferase expression (CALUX) bioassay [19]). Results from this assay will provide mechanistic support for toxicity findings.
    - Genomic evaluation in 3 diverse cell lines (e.g., HepaRG, HL60, MCF7) and subsequent pathway analysis and principal component analysis. Evaluation

[1] Parallel histology on female reproductive tissue is not proposed at this time. Although there is evidence that PAHs display female germ cell toxicity as measured by decreased follicles present in ovaries [7], that endpoint was regarded as rate limiting and is reserved for future consideration.

of the results from this *in vitro* assay and the *in vivo* gene expression results will provide support for *in vitro* to *in vivo* extrapolation.

3. Zebrafish developmental assay. This assay has been used extensively in assessing developmental and neurotoxicological effects of a wide array of chemicals [20, 21]. Specifically, work on PAHs has been conducted in this model and has yielded important hazard characterization and mechanistic data [22, 23]. Relevant endpoints that will be assessed in this model include developmental (e.g., rate of development, survival, morphology) and behavioral (e.g., motility in light and dark) endpoints.

Challenges associated with the short-term testing panel

The major challenge of the short-term testing panel is ensuring the various endpoints and assays can be clearly linked to human-relevant pathologies through association of early effects with late effects and responses in alternative/*in vitro* models with apical endpoints. For example, developmental toxicity as measured by changes in zebrafish morphology will not be automatically translatable to an equivalent developmental toxicity in humans. In order to strengthen the impact of the program and build bridges across *in vitro*/alternative animal models and mammalian models, it may be necessary to further explore observed patterns of PAH-mediated effects in targeted in vivo studies (e.g., performing supportive developmental toxicity assessments in a mammalian model with select PAHs).

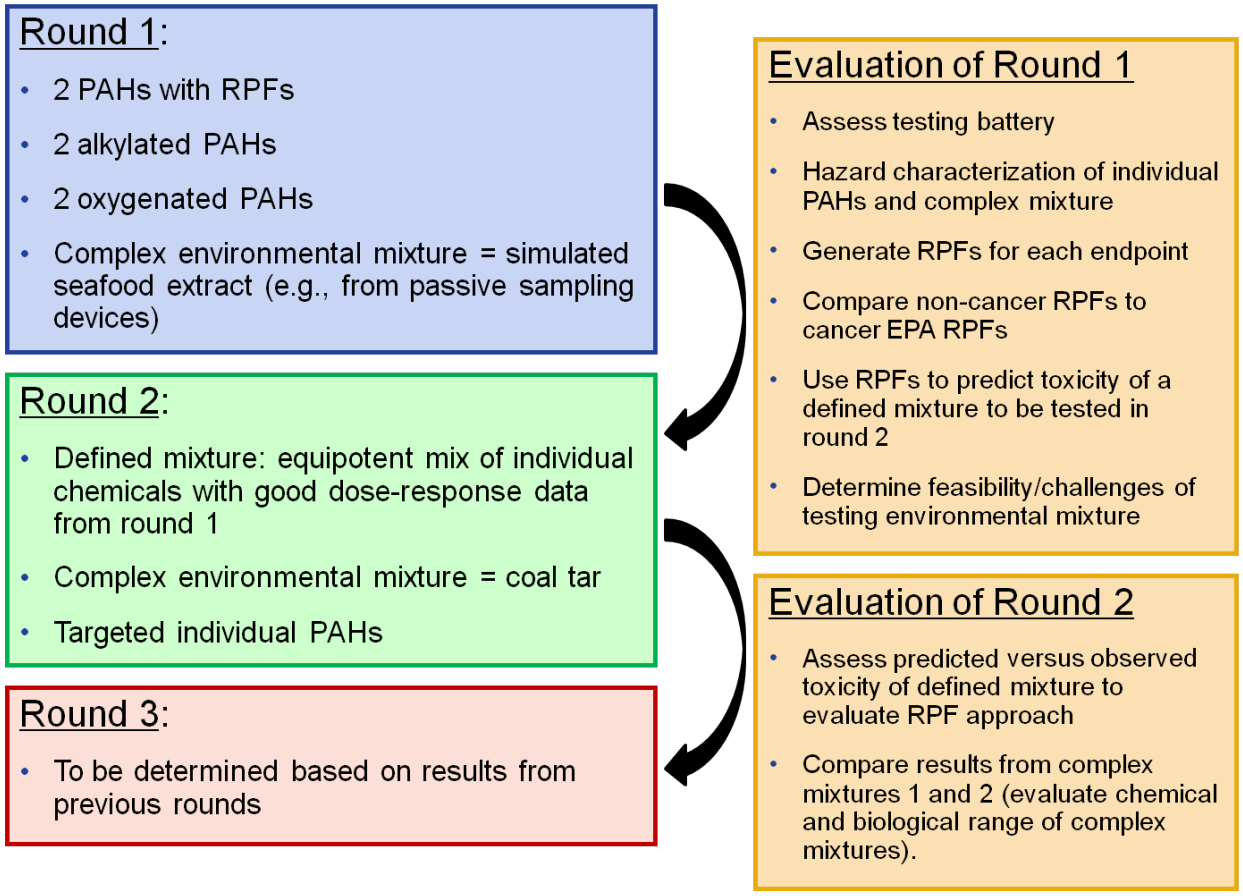
Iterative testing approach

An iterative testing approach (i.e., multiple rounds of testing) will expedite hazard characterization data for key individual PAHs, while allowing for rapid course correction to avoid unnecessary resource expenditure. Due to the complexity of the issue and the vast expanse of unknown territory, the testing program is built in units with each round informing the development of subsequent rounds. In the beginning rounds of testing, the panel of test articles will include several hazard characterization elements, as well as exploratory elements that will be used for hypotheses development. The inherent flexibility of the proposed approach will allow the project to evolve as data are generated (e.g., new assays or endpoints may replace original ones). A schematic of the iterative approach is provided below (Figure 2).

As presented in Figure 2 below, the first round of testing could include assessment of select individual PAHs from various subcategories (unsubstituted, alkylated, oxygenated), as well as a complex environmental mixture, in the short-term testing panel described above. Following completion of round 1 testing, these data will be evaluated to inform various knowledge gaps. First, the short-term testing panel will be examined. Results will be compared across endpoints to determine which measures are most informative, reliable, and sensitive. If necessary, adjustments to the panel will be made in subsequent testing rounds. Second, hazard characterization data will be gathered for the individual PAHs and the complex mixture. Next, relative potency factors will be generated for each individual PAH. These could be used to design an equipotent mixture that could be tested in the second round. Following the evaluation of results from round 1 testing, another set of individual chemicals and mixtures will be

prioritized for testing in a second round. This process of testing, evaluation, and design will then be repeated.

Figure 2. Schematic describing the proposed iterative approach with examples of possible individual PAHs and PAH mixtures for testing.



Challenges associated with the iterative approach

Although the iterative approach affords the opportunity to adjust assays and modify the direction of testing, it also has the potential to complicate comparisons across testing rounds. The impact of testing of multiple chemicals in a multi-round paradigm will be minimized through the strategic incorporation of a reference compound for standardization. Furthermore, the impact of the program will increase with the number of rounds completed and the number of assays consistently generating useful data. Minimally, completing one round of testing will provide dose-response data for multiple unknown PAHs and a relevant environmental mixture.

Cross-disciplinary collaboration strategy

In order to maximize the impact of the outlined testing strategy, steps will be taken to leverage the expertise of intramural and extramural scientists engaged in similar research. Mechanisms for accomplishing this include, but are not limited to:

- A Division of Extramural Research and Training (DERT) representative will participate in study design meetings and act as a liaison to DNTP.
- Extramural exposure scientists will be consulted during selection of complex mixtures for testing.
- Options for making materials resulting from the testing program (e.g., study samples) available to extramural partners interested in performing additional analyses will be investigated.
- Potential collaborations with complementary extramural PAH researchers will be explored.
- Continued discussion of lessons learned and suggested paths forward will be conducted with Health Canada, EPA, FDA, and other partners.

Significance and Expected Outcome

The studies proposed in this concept should contribute data and better understanding to three major areas: (1) characterizing the toxicity of PAHs, (2) building bridges between alternative animal/*in vitro* and *in vivo* studies, and (3) developing and refining predictive models of mixture toxicity. The strengths of the proposed testing program include: (1) a focus on an important class of environmentally relevant compounds, (2) broad coverage of toxic endpoints, (3) a comparison of a significant number of individual PAHs and PAH-containing mixtures using a consistent experimental paradigm, and (4) deliverable units of data throughout the iterative program (i.e., each round provides useful information and does not depend on the results from the next round for success).

References

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