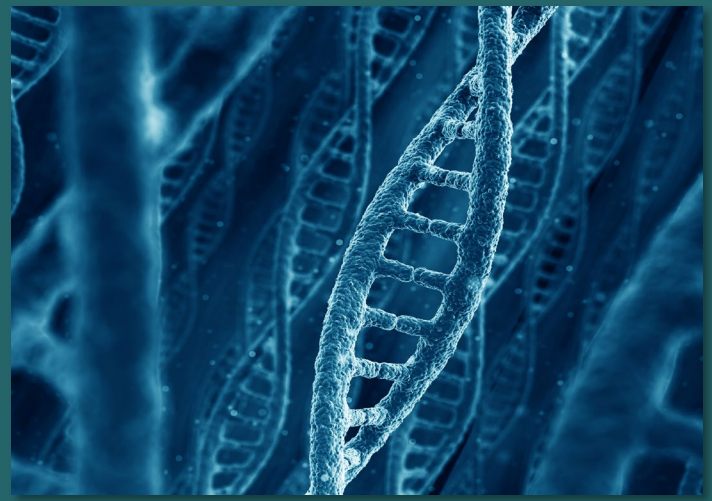
Scientific Support for ETAP: Introduction

Alison Harrill, Ph.D. – Associate Director for Toxicology



The views expressed in this presentation are those of the presenter and do not necessarily reflect the views or policies of the U.S. EPA

Objectives

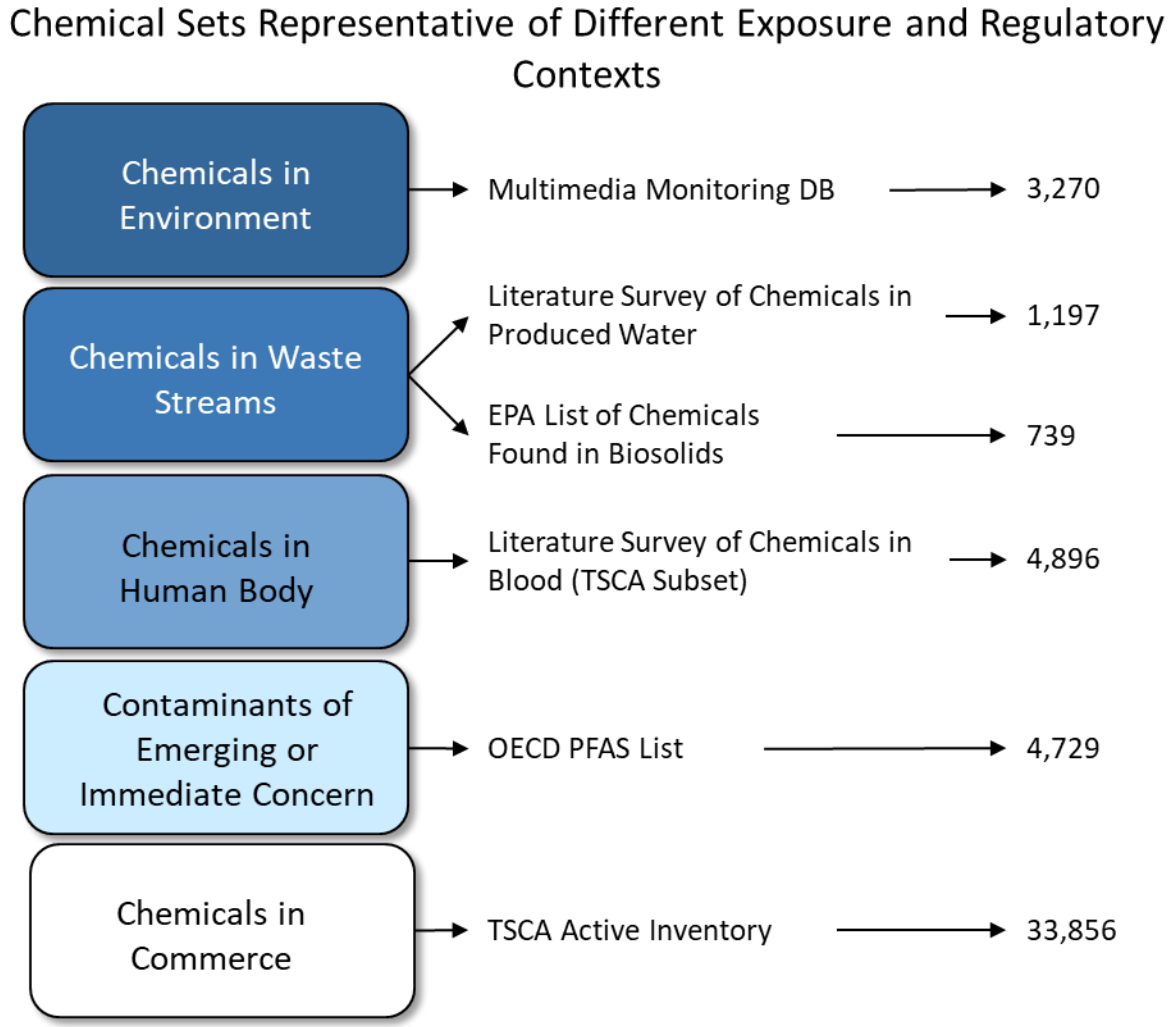
- Provide a high-level overview of the need for an additional ORD assessment product
- Worldwide and domestic chemical and toxicity testing and human health assessment landscape
- EPA history, policies, use of transcriptomics, technical and enabling advances

Chemical Landscape

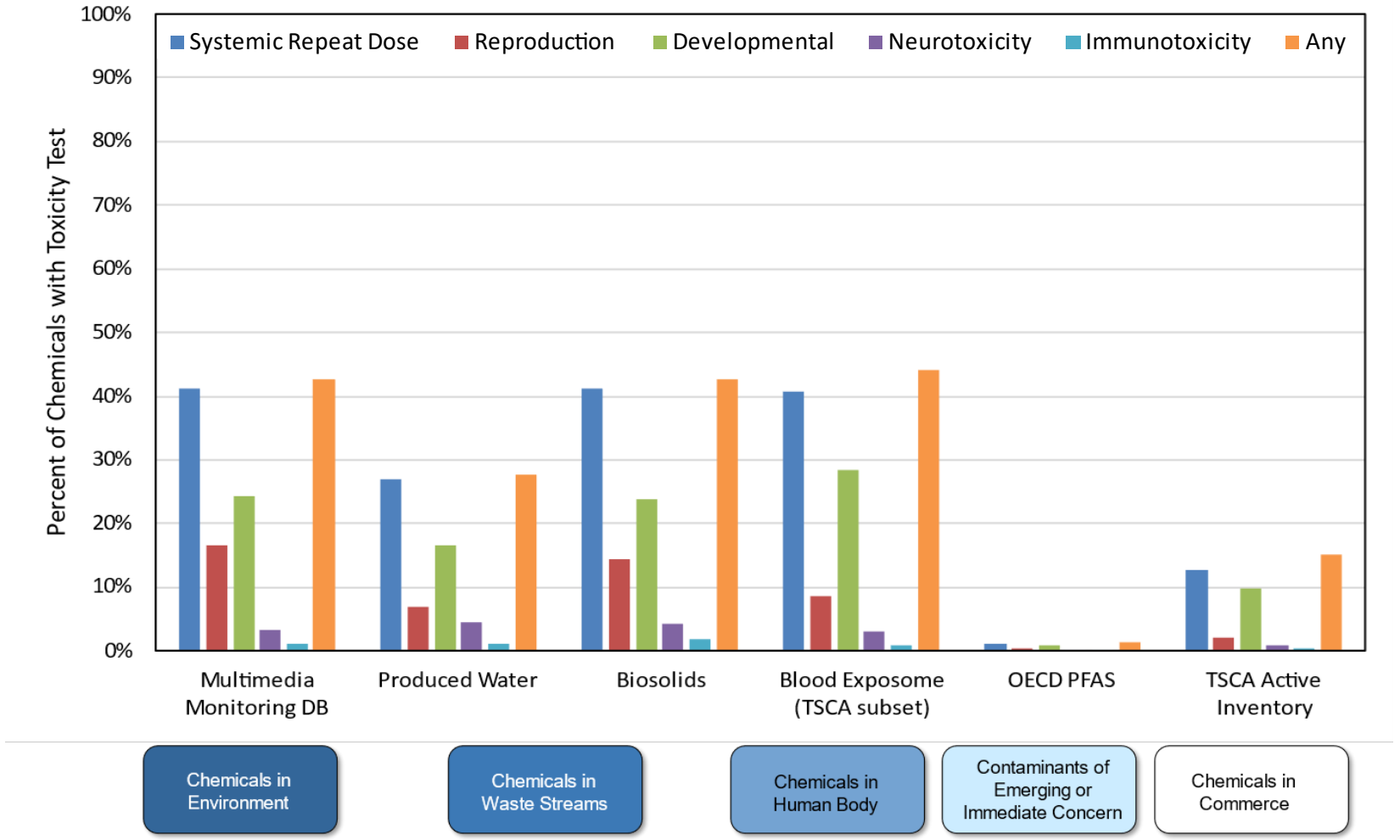
- >95% of manufactured goods and articles are estimated to be reliant upon an industrial chemical process (*Oxford Economics* 2019)
- >350,000 chemicals or mixtures registered in one or more inventories among 19 countries and regions (Wang et al. *Environ Sci Technol* 2020)
- In US, TSCA inventory contains >86,000 chemicals, with 42,000 commercially active
- These numbers are a snapshot in time, trends in chemical production continue to rise

Toxicity Testing Landscape

- Understanding human health impacts of chemical exposures requires testing and access to toxicity data, traditionally animal data
- Entails guideline animal studies: general acute, subchronic, chronic repeated dose toxicity studies and special studies, including neuro-, repro-, immuno-, and developmental toxicity
- Requirements for testing vary depending on intended use of the chemical and relevant statutes



% of List Chemicals with Available Toxicity Data



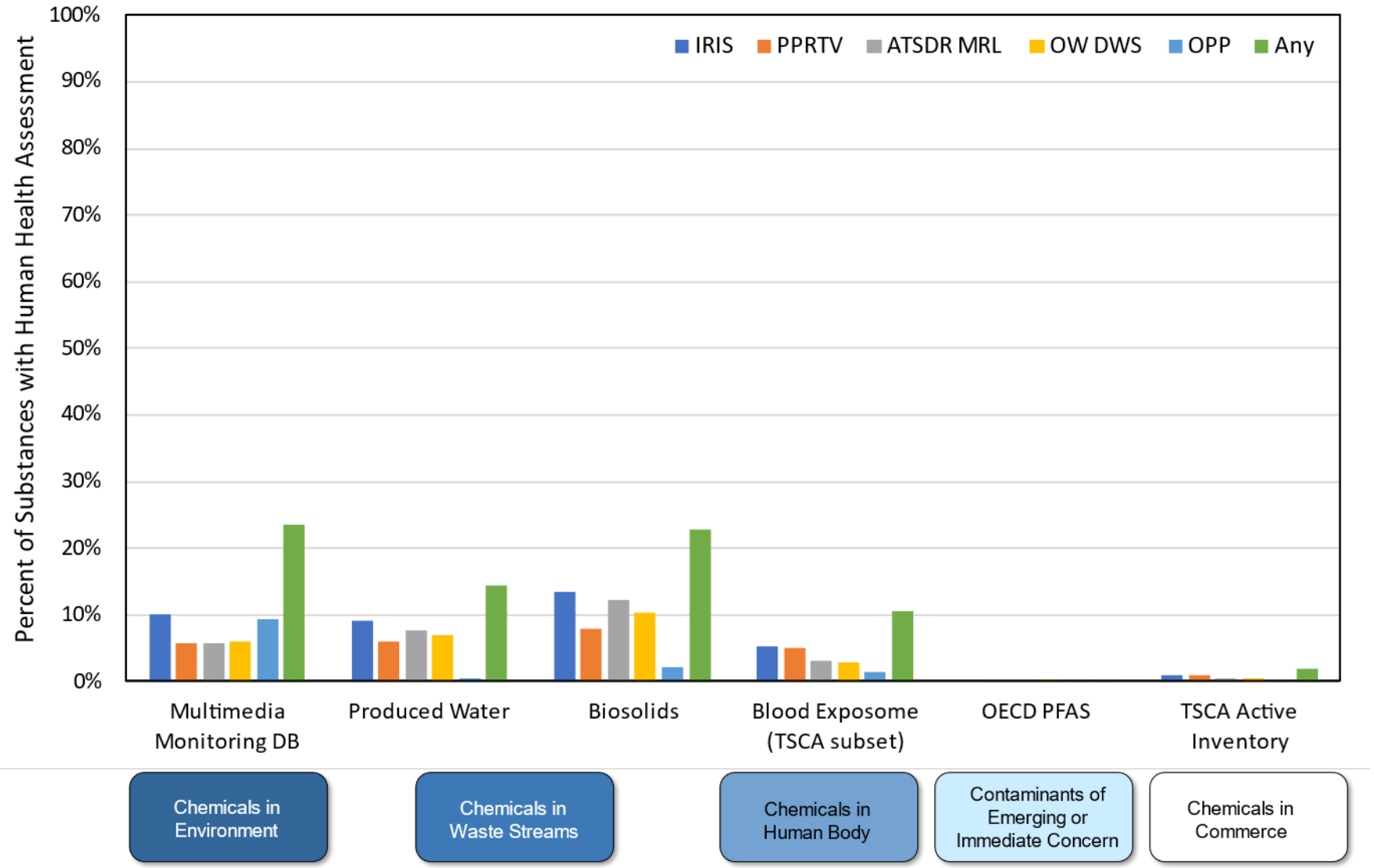
The sets of chemicals were selected to represent substances found in environmental media (Multimedia Monitoring Database), different waste streams (produced water from oil and gas extraction; biosolids), the human body (TSCA subset of the blood exposome), contaminants of emerging concern (OECD PFAS list), and commerce (TSCA active inventory). The 'Systemic Repeat Dose' toxicity test includes repeat dose studies of subchronic and chronic duration. The 'Any' category is the union of unique chemicals across the various study types. The total percentages of chemicals across study types may not equal the total percentage in 'Any' given that chemicals may have multiple different studies. The percentages of chemicals with toxicity tests were calculated based on the respective studies in ToxValDB v9.4.

Human Health Assessment Landscape

- Time required beyond testing to complete an assessment is substantial
  - 15-36 months to review testing results of a conventional agricultural pesticide following data collection (FIFRA SAP 2011)
  - It can take ≥4 years to develop human health assessment for industrial and commercial chemicals (Krewski et al. 2020)
  - More complex assessments can take substantially longer (NASEM 2009)

- Insufficient data, coupled with the required time and resources has led to fewer chemicals with reference values for regulatory applications

% of List Chemicals with Human Health Assessments



IRIS – US EPA Integrated Risk Information System

PPRTV – US EPA Provisional Peer Reviewed Toxicity Values

ATSDR MRL – Agency for Toxic Substances and Disease Registry Minimal Risk Levels

OW DWS – US EPA Office of Water Health Advisories

OPP – US EPA Office of Pesticide Programs

ETAP

For data-poor chemical substances with insufficient existing or publicly accessible repeated dose experimental animal toxicity studies or suitable human epidemiological evidence

Completion target is 9 months from chemical procurement to ETAP issuance

Discussion Agenda

First session: Scientific support

Second session: ETAP process

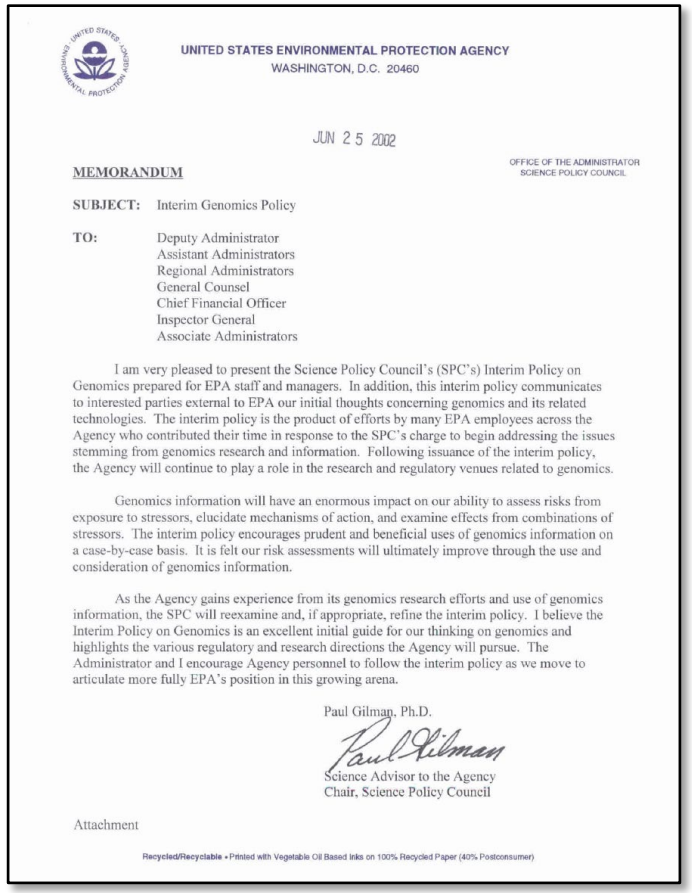
- Proposing to develop transcriptomic reference values (TRV) via EPA Transcriptomic Assessment Products
- To meet the need for toxicity testing and human health assessment of chemicals

Development of the EPA Transcriptomic Assessment Product (ETAP)

- Transcriptomics entails large-scale measurement of gene expression changes and application to toxicology enables broad characterization of biological processes impacted following chemical exposure
- Transcriptomics technology is mature, is reproducible, and has become widely available
- Detailed in subsequent talks, a substantial body of research has demonstrated that doses causing transcriptional changes are concordant with doses causing adverse apical effects in traditional tox studies, underscoring its potential application to regulatory decision making

Evolution in Application of Transcriptomics at EPA

- EPA has longstanding commitment toward utilizing emerging technologies to enhance testing paradigms and improve the utility and predictability of risk assessment methods
- Transcriptomic data are attractive because changes in gene transcript (mRNA) expression are frequently observed to precede or coincide with apical effects
- EPA released interim policy on genomics in 2002
  - Advocated using transcriptomics data on case-by-case basis in a weight of evidence approach



UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

JUN 25 2002

OFFICE OF THE ADMINISTRATOR
SCIENCE POLICY COUNCIL

MEMORANDUM

SUBJECT: Interim Genomics Policy

TO: Deputy Administrator
Assistant Administrators
Regional Administrators
General Counsel
Chief Financial Officer
Inspector General
Associate Administrators

I am very pleased to present the Science Policy Council's (SPC's) Interim Policy on Genomics prepared for EPA staff and managers. In addition, this interim policy communicates to interested parties external to EPA our initial thoughts concerning genomics and its related technologies. The interim policy is the product of efforts by many EPA employees across the Agency who contributed their time in response to the SPC's charge to begin addressing the issues stemming from genomics research and information. Following issuance of the interim policy, the Agency will continue to play a role in the research and regulatory venues related to genomics.

Genomics information will have an enormous impact on our ability to assess risks from exposure to stressors, elucidate mechanisms of action, and examine effects from combinations of stressors. The interim policy encourages prudent and beneficial uses of genomics information on a case-by-case basis. It is felt our risk assessments will ultimately improve through the use and consideration of genomics information.

As the Agency gains experience from its genomics research efforts and use of genomics information, the SPC will reexamine and, if appropriate, refine the interim policy. I believe the Interim Policy on Genomics is an excellent initial guide for our thinking on genomics and highlights the various regulatory and research directions the Agency will pursue. The Administrator and I encourage Agency personnel to follow the interim policy as we move to articulate more fully EPA's position in this growing arena.

Paul Gilman, Ph.D.
Science Advisor to the Agency
Chair, Science Policy Council

Attachment

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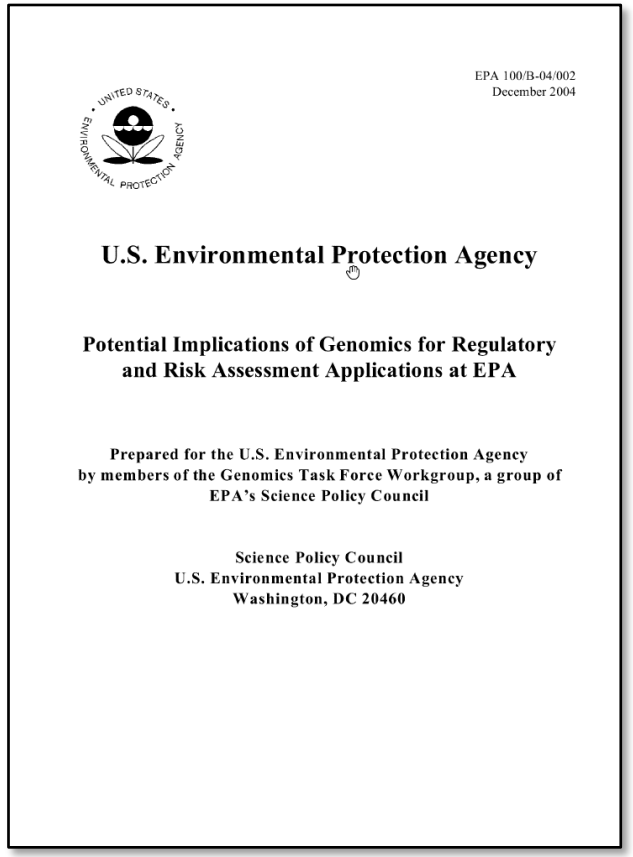
U.S. EPA, Science Policy Council, 2002.

Evolution in Application of Transcriptomics at EPA

- EPA published first report on potential applications of genomics in chemical risk assessment in 2004
- Emphasized potential applications in prioritization, monitoring, reporting provisions, mode of action, identifying sensitive populations, and addressing mixtures
- Gaps identified to be addressed: adequate technical infrastructure and training of qualified personnel, need for technical framework for data analysis, need standardization criteria for acceptance of transcriptomic data



EPA 100/B-04/002
December 2004

U.S. Environmental Protection Agency

Potential Implications of Genomics for Regulatory and Risk Assessment Applications at EPA

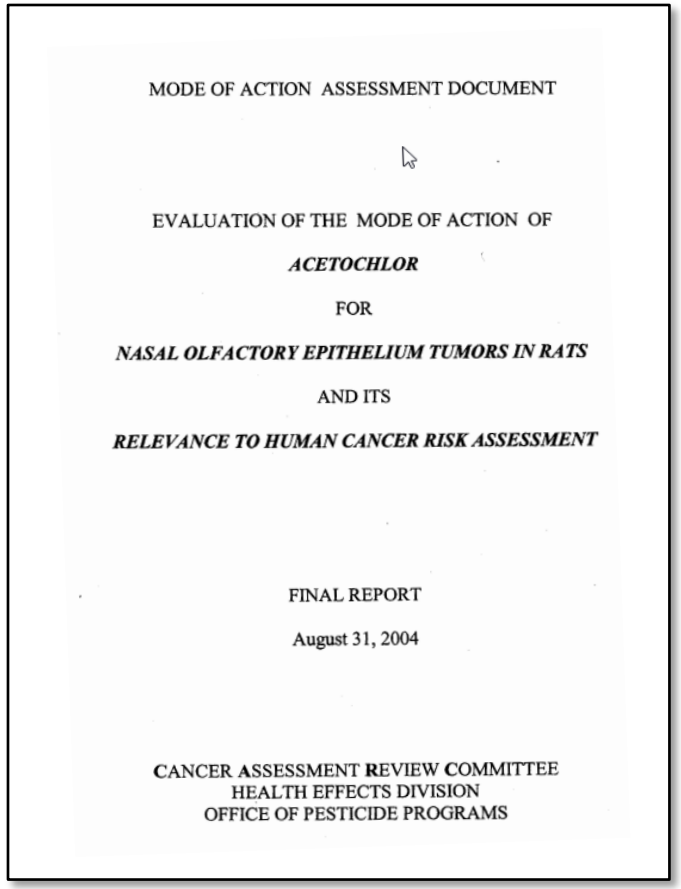
Prepared for the U.S. Environmental Protection Agency by members of the Genomics Task Force Workgroup, a group of EPA's Science Policy Council

Science Policy Council
U.S. Environmental Protection Agency
Washington, DC 20460

U.S. EPA, Science Policy Council, 2004.

Evolution in Application of Transcriptomics at EPA

- EPA used transcriptomics information in a mode-of-action weight of evidence cancer risk assessment in 2004
- Time course transcriptomics data was derived from rat olfactory mucosa at a single dose
- Early gene expression changes interpreted to be consistent with oxidative damage to DNA followed by cell proliferation, while late gene expression changes interpreted to be consistent with tumorigenic progression



MODE OF ACTION ASSESSMENT DOCUMENT

EVALUATION OF THE MODE OF ACTION OF

ACETOCHLOR

FOR

NASAL OLFACTORY EPITHELIUM TUMORS IN RATS

AND ITS

RELEVANCE TO HUMAN CANCER RISK ASSESSMENT

FINAL REPORT

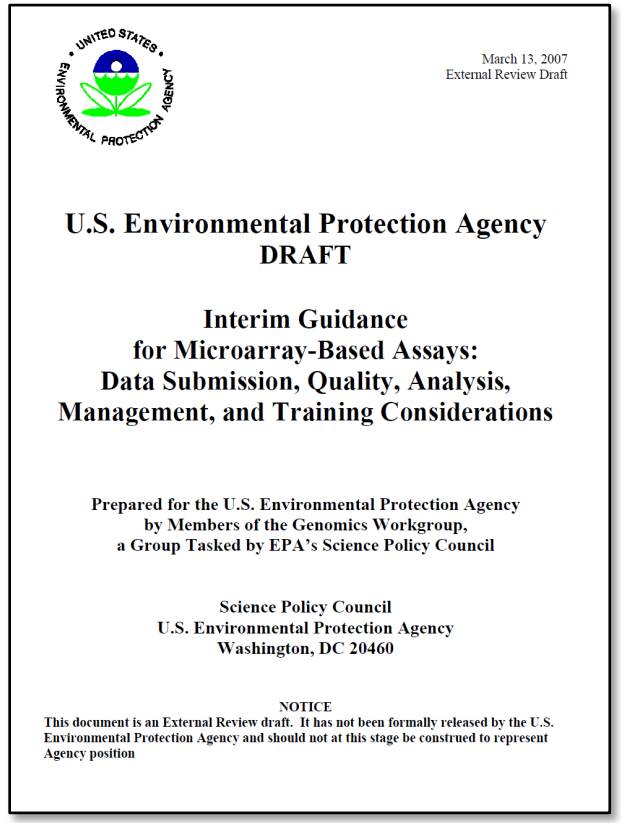
August 31, 2004

CANCER ASSESSMENT REVIEW COMMITTEE
HEALTH EFFECTS DIVISION
OFFICE OF PESTICIDE PROGRAMS

U.S. EPA, Office of Pesticides Programs, 2004.

Evolution in Application of Transcriptomics at EPA

- EPA released interim guidance for microarray data submissions, quality, and analysis in 2007
- Provided recommendations on performance approaches for quality assessment parameters, data analysis, Agency data submissions, and data management
- Issued a draft Genomics Data Evaluation Record template
- Recommended development of
  - Training modules and materials for risk assessors,
  - Cross-Agency collaboration, and
  - Case study application



March 13, 2007
External Review Draft

U.S. Environmental Protection Agency
DRAFT

Interim Guidance for Microarray-Based Assays: Data Submission, Quality, Analysis, Management, and Training Considerations

Prepared for the U.S. Environmental Protection Agency by Members of the Genomics Workgroup, a Group Tasked by EPA's Science Policy Council

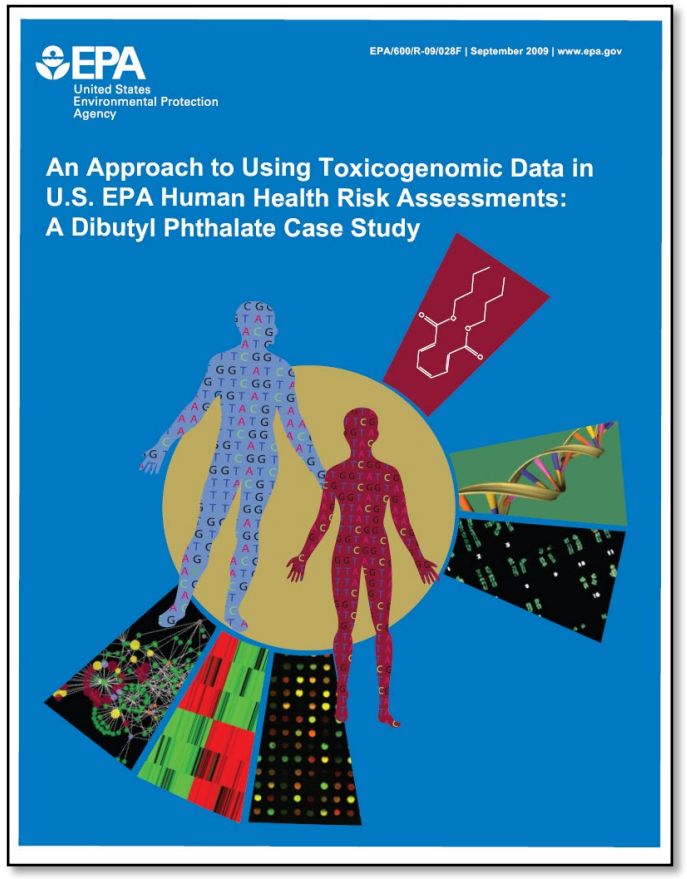
Science Policy Council
U.S. Environmental Protection Agency
Washington, DC 20460

NOTICE
This document is an External Review draft. It has not been formally released by the U.S. Environmental Protection Agency and should not at this stage be construed to represent Agency position

U.S. EPA, Science Policy Council, 2007.

Evolution in Application of Transcriptomics at EPA

- EPA released a case study for application of transcriptomic data to human health risk assessment in 2009
- Outlined a systematic and flexible approach to accommodate different health and risk assessment practices
- Focused primarily on informing mode-of-action as part of a weight-of-evidence
- Provided some recommendations on best practices and highlighted current limitations



U.S. EPA, Office of Research and Development, 2009.

Reproducibility of Transcriptomic Data

- Confidence in application of transcriptomics data has been bolstered by international consortia
- MAQC (led by FDA) in cross-site, cross-platform studies demonstrated strong inter- and intra-platform reproducibility using RNA reference samples (MAQC 2006)
- SEQC evaluated next-gen sequencing tech, establishing best practices for RNA-seq methods (SEQC 2014), characterizing inter-platform reproducibility of RNA-seq protocols and technologies (Li et al. 2014), and evaluating bioinformatic tools. SEQC cross-site, cross-platform studies demonstrated high consistency in RNA-seq results with intra- and inter-platform Spearman rank concordance values at 0.86 and 0.83

Reproducibility of Transcriptomic Data

- SEQC2 (Mercer et al. 2021): 300 scientists from 150 industry, academic, government organizations
- Evaluated a variety of NGS technologies, including RNA-seq for germline variant detection, cancer genomics, biomarker discovery, precision medicine
- SEQC2 results provided a scientific foundation that enabled regulatory approvals of NGS-based liquid biopsy tests for solid tumors as companion diagnostics
- Increased confidence in use of transcriptomic-based technologies for regulatory applications

Transcriptomics Data Analysis Infrastructure

- Software for transcriptomic dose response developed via intergovernmental partnership (BMDExpress2)
  - Employs validated continuous parametric models deployed in EPA's BMDS software
  - Curve fits computed for each transcript, followed by functional classification analysis that assigns transcriptomic features into pre-defined gene sets (GO) and determines gene set potency estimates as BMD(L)
  - Methods and assumptions reviewed by NIH-assembled panel of experts (NTP 2018)

Development of Omics Reporting Template - OECD

- The Organization for Economic Cooperation and Development (OECD) introduced an internationally accepted transcriptomic reporting framework in 2021
  - Provides guidance on reporting of transcriptomic information that fosters transparency and reproducibility
  - Captures experimental information, data acquisition/processing, and data analysis
  - Ensures sufficient information is available to enable evaluation of experimental data and interpretation
- Template is awaiting formal approval at the OECD and – once adopted – will be deployed for agency usage and the ETAP

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Commentary

Progress towards an OECD reporting framework for transcriptomics and metabolomics in regulatory toxicology

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J Harrill *et al.* Reg. Toxicol. Pharmacol. , 2021

Infrastructure Enabling the ETAP

- Addressed prior gaps analyses - EPA has sufficient staff, training, and infrastructure to conduct transcriptomic studies and analyze the data
- Experience in utilizing transcriptomics data to support human health assessments
- Reliable and reproducible wet lab and data analysis workflows
- Collaborations with domestic and international partners led to development of robust reporting frameworks for regulatory review
- Tested software and dose response analytic workflows with expert review (Scott)
- Extensive body of literature that enables optimization of study designs (Leah, Logan, Kelsey)