Evolution from Traditional Data Requirements to Knowledge-based Requirements: EDSP21 Work Plan

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EDSP Targeted Mission

To protect public health and wildlife by screening and testing chemicals and taking appropriate actions for those chemicals that are found to have endocrine effects.
Endocrine Disruptor Screening Program
Legislative Mandate

- **1996 Federal Food, Drug and Cosmetic Act, section 408(p)**
  Requires the U. S. EPA to develop a screening program using appropriate validated test systems and other scientifically relevant information to determine whether certain substances may have an effect in humans that is similar to an effect produced by a naturally occurring estrogen, or other such endocrine effect as the Administrator may designate.

- **1996 Safe Drinking Water Act Amendments, section 1457**
  Testing of chemical substances that may be found in sources of drinking water, if substantial human populations may be exposed.
1998 EDSTAC Recommendations:
- Protect Human Health and Wildlife
- Include Estrogen, Androgen and Thyroid pathways
- Develop a two-tiered screening and testing program:

**Tier 1 Screening**

potential to interact with the estrogen, androgen or thyroid hormone systems

**Tier 2 Testing**

if endocrine-mediated adverse effects then quantify dose-response
### EDSP Tier 1 Screening Battery

<table>
<thead>
<tr>
<th><strong>In vitro</strong></th>
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<tbody>
<tr>
<td>Estrogen Receptor (ER) Binding</td>
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<tr>
<td>Estrogen Receptor Transcriptional Activation Assay (ERTA)</td>
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<tr>
<td>Androgen Receptor (AR) Binding</td>
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<td>Steroidogenesis</td>
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<td>Aromatase</td>
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<tr>
<td><strong>In vivo</strong></td>
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<tr>
<td>Uterotrophic (rat)</td>
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<td>Hershberger (rat)</td>
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<td>Pubertal Female (rat)</td>
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<td>Pubertal Male (rat)</td>
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<td>Amphibian Metamorphosis Assay (frog)</td>
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<td>Fish Short-Term Reproduction Assay</td>
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<td>Steroidogenesis (H295R)</td>
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<td>Aromatase (Recombinant)</td>
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<td>Fish Reproductive Screen</td>
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<td>Amphibian Metamorphosis</td>
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## Proposed EDSP Tier 2 Tests

<table>
<thead>
<tr>
<th>Category</th>
<th>Description</th>
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</table>
| Mammalian Two-Generation Reproduction | (Sprague Dawley rat)  
( mamma may be replaced by Extended F1-Generation) |
| Avian Two-Generation Reproduction | (Japanese quail) |
| Larval Amphibian Growth and Development | (Xenopus laevis) |
| Fish Multi-Generation Reproduction | (Medaka) |
| Invertebrate Multi-Generation Reproduction | (Mysid and Copepod) |
The Future of Toxicology

...the application of mathematical and computer models and molecular biological approaches to improve...prioritization of data requirements and risk assessments.

To support the evolution of toxicology from a predominantly observational science at the level of disease-specific models to a predominantly predictive science focused upon a broad inclusion of target specific, mechanism-based, biological observations.

...a new toxicity-testing system that evaluates biologically significant perturbations in key toxicity pathways by using new methods in computational biology and a comprehensive array of in vitro tests based on human biology.
Tox21 Vision

Transition 21st century technologies, to enhance the efficiency and effectiveness of chemical risk management.

**CURRENT**

Heavy reliance on animal studies

Generate information for all possible outcomes

Based on traditional toxicity tests

**FUTURE**

Less reliance on animal studies

Tailor data generation

Based on understanding of toxicity pathways
Less reliance on animal testing; more knowledge-based instead.

More detailed *in vitro* assays, enhanced exposure assessment, greater specificity of *in silico* models.

Greater certainty necessitates increased understanding, quantitative data, and greater integration at each level.
Adverse Outcome Pathway Concept

- Key to achieving goal.
- Framework that links the direct molecular initiating event to an adverse outcome at a level of biological organization relevant to risk assessment.
- Basis for
  - Integrating lower tier tests and non-animal models
  - Applying read across methods
  - Development of Expert Systems
- Consistent with Mode of Action analysis
Application to Levels of Organization Based on Source to Outcome

Source

Environmental Contaminant

Exposure

Molecular Initiating Event

Cellular Effects

Source to Outcome Pathway

Adverse Outcome Pathway

Mode of Action

Toxicity Pathway

Community

Population

Individual
Adverse Outcome Pathway and the Data Streams that Inform the Pathway

Adverse Outcome Pathways

Structure Activity Relationships

In vitro studies

In vivo studies

Toxicity Pathways

Chemicals → Pharmaco-kinetics ↔ Molecular Target ↔ Cellular Response ↔ Tissue/Organ

Individual ↔ Population

Biomonitoring data
**Biologic inputs**

- Exposure
- Uptake-Delivery to Target Tissues
- Perturbation

**Cellular response pathway**

- Biologic inputs
- Early cellular changes
- Adaptive Responses

**Toxicity Pathway**

- Molecular initiating event
- Perturbed cellular response pathway

**Adverse Outcome Pathway**

- Adverse outcome relevant to risk assessment
- Cell injury, inability to regulate
- Adverse outcomes (e.g., mortality, reproductive impairment)

"Normal" Biological Function

**Adverse Outcome (e.g., mortality, reproductive impairment)**
EDSP21 Work Plan Summary
(USEPA, September 2011)

www.epa.gov/endo
EDSP21 Objective

- Maximize use of existing data.
- Targeted *in vivo* toxicity screening.
- Use a variety of tools in a tiered testing and assessment framework.
- Systematically and *incrementally* incorporate new tools, methodologies.
- Advance understanding of key events in toxicity pathways.
The EDSP21 Work Plan describes: Multi-level and Integrated approach to determine whether a chemical has the potential to interact with E, A, or T.

Three main objectives:
(1) Prioritization - The near-term goal (<2 years)
(2) Screening - The intermediate-term goal (2-5 years)
(3) Data Replacement – The long-term goal (>5 years)
EPA Research provides basis for improving the suite of assays and models to advance chemical prioritization and screening.

The universe of chemicals passes through each version of the HTP/in silico pipeline to evaluate chemicals in refined tests, for new pathways, to evaluate, improve, and validate methods.

**Chemicals Of Regulatory Interest**

- **Near-Term (<2 yrs)**
  - In vitro HTP/ in silico
  - Test+ -> Current EDSP T1S Battery -> WOE+

- **Intermediate –Term (2-5 yrs)**
  - In vitro HTP/ in silico
  - Test+ -> in vitro/in silico to focus subset of EDSP T1S
  - WOE+

- **Longer-Term (>5 yrs)**
  - In vitro HTP/ in silico (full replacement of Tier 1)
  - WOE+
EDSP21 Work Plan

Pre-Prioritization
Develop science-based policy and tools for prioritization and screening
- Establish EDSP21 work group across EPA
- Develop and establish reference chemical library and EDSP21 database
- Develop a prioritization process with criteria to determine order for screening universe of non pesticide active ingredient chemicals
- Develop a validation process to evaluate in silico and in vitro HTP methods for screening
- Develop and evaluate exposure model

Intermediate-term goal: incorporate computational or in silico models and molecular-based in vitro high-throughput (HTP) screening assays into EDSP Tier 1 screening.

Short-term goal: use computational or in silico models and molecular-based in vitro high-throughput (HTP) screening assays to prioritize chemicals for EDSP Tier 1 screening.

Prioritization
Determine the order of chemicals for screening
- Identify the universe of chemicals
- Identify in silico models and in vitro HTP assays for EAT
- Prioritize based on re-registration, existing exposure and effects information, and results from in silico and in vitro HTP methods
- Establish list of chemicals for screening
- Send orders for Tier 1 screening to determine potential to interact with E, A, or T

Screening
Optimize Tier 1 screening to determine the need for Tier 2 testing
- Compare EAT information from prioritization to results of Tier 1 screening for E, A or T
- Utilize Tier 1 screening results as a step in validation process for using HTP assays to screen for the potential to interact with E, A or T
- Integrate valid HTP assays into Tier 1 screening
- Allow for public comment and peer review before regulatory acceptance
Chemical Prioritization

• Consideration of multiple data streams
  – HTP assays for estrogen, androgen and thyroid
  – Inherent chemical properties
  – Modeling predictions (e.g., QSAR and expert systems)
  – Data from structural analogs (read across)
  – Toxicity pathway based and anchored by biological mechanistically based understanding

*Figure taken from 1996, Chemical Manufacturers Association Product Risk Management Strategy Overview
OECD (Q)SAR Validation Principles

- Defined Endpoint
- Unambiguous Algorithm
- Defined Domain of Applicability
- Appropriate Measures of Goodness-of-fit, Robustness and Predictivity
- Defined Biological Mechanism of Action, if possible
Key considerations for implementation of EDSP21

- Ensure clarity of programmatic goal
- Define application and regulatory decision contexts
- Build transparent strategy with sound scientific basis
- Determine scientific validity
- Ensure public outreach
Evolution of Computational Tools

The transition from traditional empirical data to computational tools must evolve slowly in incremental steps, with strong confidence and adequate assurance that no single apical health endpoint will be left behind.