Introduction to Adverse Outcome Pathways and the AOP Wiki

Sunday July 20, 2017
10:00 AM - 12:00 PM
Sheraton Seattle, Aspen Room

10:00: Introduction to the OECD AOP Programme and Online Training course
Kate Willett, Human Toxicology Project Consortium

10:40: Building AOPs for Neurotoxicity: Perspective from an Academic
Prof. Dr. Ellen Fritsche, IUF – Leibniz Research Institute for Environmental Medicine

11:20: Demonstration and Hands-On Activity with AOP Wiki
Kristie Sullivan, Physicians Committee for Responsibility Medicine
Introduction to the OECD AOP Programme and Online Training course

Catherine Willett, Humane Society of the United States, Humane Society International
Outline: Adverse Outcome Pathways

- **Why**
  - Need for faster, predictive approach to toxicology
  - Need for better access/organization of existing and future data

- **What**
  - Purpose, definition
  - OECD AOP Program

- **How**
  - AOP Wiki
  - Guidance
  - Evaluation

- **When**
  - Use in decision making
  - Support of integrated Approaches to Testing and Assessment
Need for faster, predictive toxicology

“Transform toxicity testing from a system based on whole animal testing to one founded primarily on *in vitro* methods that evaluate changes in biologic processes using cell, cell lines, or cellular components, preferably of human origin.”

Including “tests that assess critical mechanistic endpoints involved in the induction of overt toxic effects rather than the effects themselves.”

National Academy of Sciences, 2007
Need for better access to and organization of data

Data in published papers (pdfs), reports, report summaries are not suited to:

- Facilitating collaboration and crowd-sourcing
- Avoiding duplicative effort
- Integration and analysis
- Searching and machine-reading

Adapted from D. Villeneuve
Adverse Outcome Pathways: linking molecular initiation to adverse outcomes

How to use molecular understanding to make better decisions about chemical safety
“Conceptually, an AOP can be viewed as a sequence of events commencing with initial interactions of a stressor with a biomolecule in a target cell or tissue (i.e., molecular initiating event), progressing through a dependent series of intermediate events and culminating with an adverse outcome.”

“AOPs are typically represented sequentially, moving from one key event to another, as compensatory mechanisms and feedback loops are overcome.”

AOP Provides Understanding & Scaffold for Data

- Toxicity Pathways
  - Toxicants
  - Macro-Molecular Interactions
  - Cellular Responses
  - Organ Responses
- Regulatory Endpoints
  - Organism Responses
  - Population Responses

High Throughput Tox

Guideline Studies

Mechanistic Toxicology Data Bioindicators (e.g. Molecular Epi)

Epidemiology Eco Field Studies

“Borrowed” from Steve Edwards
Essential Elements of an AOP

- **Molecular Initiating Event (MIE):** Initial point of chemical interaction
- **Adverse Outcome (AO):** Adverse outcome of regulatory significance
- **Key Events (KEs) - nodes**
  - Change in biological state
  - Measurable and essential for progression
- **Key Event Relationships (KERs) - edges**
  - Connections between two key events
  - Critical for assembling evidence in support of the AO

Building an AOP

- Start anywhere
- Gather all existing knowledge
- Evaluate and document the information
- Translate and capture information as a pathway

AOP Knowledgebase: information storage, evaluation, and linkage
AOP Knowledgebase: information storage, evaluation, linkage, and modeling
AOP Wiki: information storage, evaluation, and linkage

- Captures and organizes all information and supporting documentation for KEs and KERs
- Supports OECD review and endorsement of formal AOPs
- Quantitative information is written in appropriate sections
- Not computational

Publically accessible since September 2014
www.aopwiki.org
Effectopedia

- "Explicitly captures quantitative information"
- Supports OECD review & endorsement of quantitative AOPs
- Quantitative information is intrinsic, ultimately also code execution
- 2017
OECD AOP Development Programme

What is an Adverse Outcome Pathway (AOP)
In 2012, the OECD launched a new programme on the development of Adverse Outcome Pathways. An Adverse Outcome Pathway (AOP) is an analytical construct that describes a sequential chain of causally linked events at different levels of biological organisation that lead to an adverse health or ecotoxicological effect (see figure). AOPs are the central element of a toxicological knowledge framework being built to support chemical risk assessment based on mechanistic reasoning.

Extended Advisory Group for Molecular Screening & Toxicogenomics (EAGMST)
- Guidance & Training
  - Guidance, User Handbook, many training options
- International Knowledgebase to capture information
- >100 AOPs at various stages of development

Task force on Hazard Assessment (TFHA)
- Use of AOPs in regulatory decision making
- Integrated Approaches to Testing and Assessment (IATA)

OECD AOP Development Programme

- Society for the Advancement of AOPS
- Not officially part of the OECD programme
- Any person active in developing an AOP in the wiki can join
- Is another way to enter the AOP wiki
- Not necessary to make an official submission to OECD
- Good way to begin preliminary/putative AOPs
- www.saaop.org
AOP Wiki Access: three levels

1. Anyone can access the wiki, search and read entries

2. To leave comments, you will need an account
   Request an account through
   www.aopwiki.org or www.saaop.org

3. To gain write access
   Request write access the same way
   You should have a familiarity with the wiki and desire to build an AOP
Work Process for Development and Review of AOPs through OECD

1. Enter AOP Wiki through SAAOP
2. SAAOP gardeners check format and compliance with OECD guidance
3. Enter AOP Wiki through EAGMST
4. Content and structural review by EAGMST
5. Technical review by OECD expert group
6. Process review by OECD National Coordinators (WNT)
7. Publication on website in OECD Series on Adverse Outcome Pathways
AOPs are modular
- KEs and KERs are shared by multiple AOPs
- No need to re-write the same descriptions over and over
- Reusability (best practices)

AOPs are living documents
- KE and KER descriptions can be expected to evolve over time
- As descriptions are updated and expanded – all AOP descriptions they link to update automatically

AOP networks for prediction
- Entry of structured information in KB allows for de-facto assembly of AOP networks.

AOP-KB supports principles of AOP development
AOP WIKI: information storage and evaluation
Welcome to the Collaborative Adverse Outcome Pathway Wiki (AOP-Wiki)

This wiki represents a joint effort between the European Commission - DG Joint Research Centre (JRC) and U.S Environmental Protection Agency (EPA). This serves as one component of a larger OECD-sponsored AOP Knowledgebase (AOP-KB) effort and represents the central repository for all AOPs developed as part of the OECD AOP Development Effort by the Extended Advisory Group on Molecular Screening and Toxigenomics. The other major components of this knowledgebase are Effectopedia, produced by the Organisation for Economic Co-operation and Development (OECD), the AOP Xplorer, produced by the US Army Corps of Engineers - Engineering Research and Development Center, and the Intermediate Effects DB produced by the JRC. All AOPs from the AOP Knowledgebase are available via the e.AOP Portal, which is the primary entry point for the AOP-KB.

This wiki is based upon the Chemical Mode of Action wiki developed by the EPA under the auspices of the WHO International Programme on Chemical Safety (IPCS) Mode of Action Steering Group.

Disclaimer

The content of this wiki is the sole responsibility of the individual contributors and does not necessarily represent the views of the authors' organizations nor the organizations responsible for development of the AOP-Wiki or the AOP-KB. Mention of trade names or commercial products does not constitute endorsement by any of these organizations.
# AOP WIKI: search “liver fibrosis”

## AOP Title Search Results

<table>
<thead>
<tr>
<th>Id</th>
<th>Title</th>
<th>Point of Contact</th>
<th>Author Status</th>
<th>SAAOP Status</th>
<th>MIE</th>
<th>AO</th>
<th>OECD Status</th>
<th>OECD Project</th>
</tr>
</thead>
<tbody>
<tr>
<td>38</td>
<td>Protein Alkylation leading to Liver Fibrosis</td>
<td>Brigitte Landesmann</td>
<td>Open for citation &amp; comment</td>
<td>Included in OECD Work Plan</td>
<td>Protein alkylation</td>
<td>liver fibrosis</td>
<td>TFHA/WNT Endorsed</td>
<td>1.14</td>
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</table>

## AOP Fulltext Search Results

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<td>liver fibrosis</td>
<td>TFHA/WNT Endorsed</td>
<td>1.14</td>
</tr>
<tr>
<td>34</td>
<td>LXR activation leading to hepatic steatosis</td>
<td>Marina Goumenou</td>
<td>Under development: Not open for comment. Do not cite</td>
<td>Under Development</td>
<td>LXR</td>
<td>liver steatosis</td>
<td></td>
<td></td>
</tr>
<tr>
<td>144</td>
<td>Lysosomal damage leading to liver inflammation</td>
<td>Brigitte Landesmann</td>
<td>Under development: Not open for comment. Do not cite</td>
<td>Included in OECD Work Plan</td>
<td>Liver</td>
<td>Inflammation</td>
<td>Under Development</td>
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<tr>
<td>131</td>
<td>Aryl hydrocarbon receptor activation leading to uroporphyrina</td>
<td>AmadFarhat</td>
<td>Open for comment. Do not cite</td>
<td>Included in OECD Work Plan</td>
<td>AhR</td>
<td>uroporphyrina</td>
<td>EAGMST Under Review</td>
<td>1.7</td>
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</table>
AOP WIKI: information storage and evaluation

**OECD Handbook**
Step by step guide to AOP development


**AOP-Wiki**
Provides consistent structure based on the OECD handbook and facilitates collaborative AOP development

http://aopwiki.org/

New version of AOP Wiki available in November, 2016
### AOP WIKI: KER and AOP confidence evaluation

<table>
<thead>
<tr>
<th>Biological Plausibility: between KE upstream and KE downstream?</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>High (strong): Extensive understanding of KER</strong></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Essentiality: are downstream KEs prevented if upstream KE’s blocked?</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>High (strong): direct evidence from experimental studies</strong></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Empirical Evidence: amount, quality, consistent, inconsistent?</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>High (strong): extensive evidence for temporal, dose-response</strong></td>
</tr>
</tbody>
</table>

AOP Title

**Alkylation of DNA in male pre-meiotic germ cells leading to heritable mutations**

Short name: Alkylation of DNA leading to heritable mutations

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**Relationships Among Key Events and the Adverse Outcome**

<table>
<thead>
<tr>
<th>Event</th>
<th>Description</th>
<th>Triggers</th>
<th>Weight of Evidence</th>
<th>Quantitative Understanding</th>
</tr>
</thead>
<tbody>
<tr>
<td>DNA, Alkylation</td>
<td>Directly Leads to</td>
<td>Insufficient or incorrect DNA repair; N/A</td>
<td>Strong</td>
<td>Moderate</td>
</tr>
<tr>
<td>Insufficient or incorrect DNA repair; N/A</td>
<td>Directly Leads to</td>
<td>Mutations, Increase</td>
<td>Strong</td>
<td>Moderate</td>
</tr>
<tr>
<td>DNA, Alkylation</td>
<td>Indirectly Leads to</td>
<td>Mutations, Increase</td>
<td>Strong</td>
<td>Moderate</td>
</tr>
<tr>
<td>DNA, Alkylation</td>
<td>Indirectly Leads to</td>
<td>Heritable mutations in offspring, increase</td>
<td>Strong</td>
<td>Moderate</td>
</tr>
<tr>
<td>Mutations, Increase</td>
<td>Directly Leads to</td>
<td>Heritable mutations in offspring, increase</td>
<td>Strong</td>
<td>Moderate</td>
</tr>
</tbody>
</table>

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Carole Yauk –
https://aopwiki.org/wiki/index.php/Aop:15
AOP networks emerge as AOPs are entered into the AOP-Wiki

Key Events Shared by Multiple AOPs

Linkages Shared by Multiple AOPs

Courtesy of Dan Villeneuve
AOP Title [edit]

Aromatase inhibition leading to reproductive dysfunction (in fish)
Short name: Aromatase inhibition leading to reproductive dysfunction (in fish)

Relationships Among Key Events and the Adverse Outcome

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<th>Weight of Evidence</th>
<th>Quantitative Understanding</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aromatase, Inhibition</td>
<td>Directly Leads to</td>
<td>17beta-estradiol synthesis by ovarian granulosa cells, Reduce</td>
<td>Strong</td>
<td>Moderate</td>
</tr>
<tr>
<td>17beta-estradiol synthesis by ovarian granulosa cells, Reduction</td>
<td>Directly Leads to</td>
<td>Plasma 17beta-estradiol concentrations, Reduction</td>
<td>Strong</td>
<td>Moderate</td>
</tr>
<tr>
<td>Plasma 17beta-estradiol concentrations, Reduction</td>
<td>Directly Leads to</td>
<td>Transcription and translation of vitellogenin in liver, Reduction</td>
<td>Strong</td>
<td>Moderate</td>
</tr>
<tr>
<td>Transcription and translation of vitellogenin in liver, Reduction</td>
<td>Directly Leads to</td>
<td>Plasma vitellogenin concentrations, Reduction</td>
<td>Strong</td>
<td>Moderate</td>
</tr>
<tr>
<td>Plasma vitellogenin concentrations, Reduction</td>
<td>Directly Leads to</td>
<td>Vitellogenin accumulation into oocytes and oocyte growth/development, Reduction</td>
<td>Moderate</td>
<td>Weak</td>
</tr>
<tr>
<td>Vitellogenin accumulation into oocytes and oocyte growth/development, Reduction</td>
<td>Directly Leads to</td>
<td>Cumulative fecundity and spawning, Reduction</td>
<td>Moderate</td>
<td>Moderate</td>
</tr>
<tr>
<td>Cumulative fecundity and spawning, Reduction</td>
<td>Directly Leads to</td>
<td>Population trajectory, Decrease</td>
<td>Moderate</td>
<td>Moderate</td>
</tr>
</tbody>
</table>
AOP Online Training Course
AOP Online Training Course

Two volume course:
1. Introduction and Overview
2. AOP Wiki Training

Please run, download and share!

Download:
https://humantoxicologyproject.org/about-pathways-2/aop-online-course/

Run:
https://aopwiki.org/
AOP in context of hazard and risk assessment

Adverse Outcome Pathway, Ankley 2010, Villeneuve 2014
Integrated Approach to Testing and Assessment (IATA): OECD working definition

“a structured approach that strategically integrates and weights all relevant data to inform regulatory decisions regarding potential hazard and/or risk and/or the need for further targeted testing and therefore optimising and potentially reducing the number of tests that need to be conducted.”

Using an AOP within the context of an IATA

- AOP provides biological rationale
  - For weight-of-evidence interpretation
  - For design of integrated, iterative testing strategy
- Transparent communication of certainty
- Quantitative information allows prediction
Regulatory acceptance of IATA: specific case Defined Approaches (DA)

- Several different possibilities for combining information
- How do regulators deal with different IATA to satisfy same information request?
- Proper guidance is crucial
- DA → possibly covered by MAD?
  - (Mutual Acceptance of Data)

(J. Baroso, European Commission. 2014)
Regulatory acceptance of IATA: specific case Defined Approaches


- Guidance on development, evaluation and application of IATA
- Harmonized template for describing IATA

**Six Principles: Essential Information for Regulatory Application of an IATA**

1. A defined endpoint
2. A defined purpose
3. A description of the rationale underlying the construction of the IATA
4. A description of the individual information sources constituting the IATA
5. A description of how the individual information sources are integrated to derive the final prediction/assessment
6. A description of the known uncertainties associated with the IATA application
AOP-supported IATA example: Skin Sensitization

Skin sensitization IATA:

- Exposure?
- Molecular Initiating Event
  - In vitro skin absorption (OECD 428)
  - QSARs; Direct Peptide Reactivity Assay (DPRA; OECD 442C)
- Cellular Effects
  - h-CLAT; OECD 442E
  - KeratinoSens (OECD 442D)
  - MUSST (U-SENS) LuSens
- Organ Effects
  - Keratinocytes
- Individual Effects
  - Local Lymph Node Assay (LLNA, OECD 429)-mouse

Question to be answered:
- Screening?
- Hazard ID?
- GHS C&L?
- Sub-classification?
Regulatory acceptance of IATA: specific case Defined Approaches


Unclassified

Organisation de Coopération et de Développement Économiques
Organisation for Economic Co-operation and Development

ENVIRONMENT DIRECTORATE

JOINT MEETING OF THE CHEMICALS COMMITTEE AND THE WORKING PARTY ON CHEMICALS, PESTICIDES AND BIOTECHNOLOGY

GUIDANCE DOCUMENT ON THE REPORTING OF DEFINED APPROACHES AND INDIVIDUAL INFORMATION SOURCES TO BE USED WITHIN INTEGRATED APPROACHES TO TESTING AND ASSESSMENT (IATA) FOR SKIN SENSITISATION

Series on Testing & Assessment
No. 256

ENV/JM/MONO(2016)29

27-Oct-2016

English - Or. English
Regulatory acceptance of IATA: specific case Defined Approaches

Examples of different IATA for skin sensitization:

Nukada et al. (2013) Toxicology in Vitro 27, 609–618

Jaworska et al. (2013) Journal of Applied Toxicology

Bauch et al. (2012) Regulatory Toxicology and Pharmacology 63, 489–504
In Summary

- AOPs can support decision making at every level, and in several ways:
  - support WoE arguments
  - support ITS design
  - transparent communication of uncertainty
  - predicting outcome

- AOP Wiki is crowd-sourced, open to everyone
  - The more participation, the better it will be!
Thank you!

Catherine Willett, PhD
Director, Regulatory Testing
Risk Assessment and Alternatives
Humane Society of the United States
Humane Society International

Coordinator, Human Toxicology Project Consortium

kwillett@humanesociety.org
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