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PREDICTING MODES OF TOXIC ACTION FROM CHEMICAL STRUCTURE

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Like many of the papers in the *Environmental Toxicology and Chemistry* "Top 100" list, the development of the fathead minnow database [1] and the assignment of modes of action to the 617 chemicals therein resulted from a comprehensive research effort by a multidisciplinary team of researchers with expertise in quantitative structure–activity relationship (QSAR) modeling, chemistry, toxicokinetics, joint toxicity of chemical mixtures, and behavioral and aquatic toxicology. The fathead minnow database was the culmination of the US Environmental Protection Agency's (USEPA) QSAR research program, which spanned nearly 20 yr, a rare program because many research planning efforts typically run within a 5-yr window of time. The vision for the USEPA QSAR program was born out of necessity. With the promulgation of the Toxic Substances Control Act in 1975, agency risk assessors needed ready access to information and new innovative methods to determine or estimate the toxicity of existing chemicals and new chemicals submitted under the premanufacturing notification program. Along with the fathead minnow database, this research effort, by USEPA scientists and collaborators, resulted in the development of methods, databases, and computational tools that are key components in risk assessments today, including the Simplified Molecular Input Line Entry System (SMILES) convention for depicting chemical structure [2] and the ECOTOX database [3].

The reliability and relevance of a QSAR is based on the scientific validity and statistical rigor of the model, its applicability to a chemical or class of chemicals requiring estimation, and the relevance of the endpoint to the risk assessment question [4]. Therefore, the training set used in model development must meet similar criteria (e.g., reliability of study results, inclusion of chemical classes of relevance to the risk assessment). Historically, the fathead minnow database has been a major component of the training sets behind many QSAR modeling and structure–fragment tools in use today (e.g., the USEPA's ECOSAR and the Organization for Economic Cooperation and Development [OECD] QSAR Toolbox [5,6]). This is due in part to its transparency and public availability (see http://www.epa.gov/med/Prods_Pubs/fathead_minnow.htm and http://www.epa.gov/ncct/dsstox/sdf_epafhm.html), but its usefulness in QSAR models can mainly be credited to the strategic approach taken in the development of the database. The express purpose of the fathead minnow database was to build relevant and reliable QSAR models. Over 19 000 substances based on molecular topology and graph theory indices were plotted, and test chemicals were selected from a diversity of chemical neighborhoods [7]. This approach ensured that the database covered a wide range of structure-space, and thereby a wide range of possible modes of toxicity. All tests were conducted in the same laboratory following standard test methods. Therefore, both the dilution water and fish used were from a single source. In addition, all new breeding pairs introduced into the culture were tested to ensure consistent sensitivity in replicate bioassays. Chemicals used were of the highest purity, with all treatment concentrations measured under stringent data quality objectives. These factors minimized variability in the test results, thereby increasing confidence that variation in toxicity was related to variation in chemical structure and associated toxicological properties.

The fathead minnow database is not the only collection of high quality study results available for use in the development of reliable QSAR models (see Bradbury et al. [8] and http://www.epa.gov/ncct/dsstox/ for other model data sets). What makes the fathead minnow database unique is that, along with mortality data, the database includes results from behavioral toxicity assessments [9], studies on physiological responses in rainbow trout [10], and joint toxicity of chemical mixture tests [11]. Using a weight of evidence (WOE) approach, the data were then used to assign a mode of action (MOA) to each chemical. The MOA assignments benefited from the results of a 1988 USEPA-sponsored workshop that brought together experts in the field of toxicology. The workshop yielded a series of studies documenting the structural and physical/chemical attributes for MOAs associated with acute exposure (e.g., narcosis, uncoupling of oxidative phosphorylation, electrophilic reactivity, acetylcholinesterase inhibition, voltage-gated sodium channel modulators) [12].

The release of the fathead minnow database placed an emphasis on a more mechanistic approach to model development, and the OECD has subsequently identified the linkage of molecular descriptors used in QSAR models to MOA as 1 of the 5 principles for validating QSAR models [13]. Knowledge of the pathway, from the interaction of the chemical with the molecular target, to the response in the whole organism, is critical in the risk assessment arena as well [14]. Risk assessments have traditionally relied on data from apical endpoints (e.g., mortality, growth, reproduction), but as research shifts toward testing strategies focused on in vitro methods, it will be important to have a clear understanding of how

All Supplemental Data may be found in the online version of this article. See Table S1 for the number of citations and rank of all the "Top 100" papers, which in this essay is reference [1].

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these new endpoints at the lower levels of biological organization relate to the response in the whole organism. The adverse outcome pathway (AOP) has recently been proposed as a framework to organize existing knowledge associated with the molecular initiating event at the target site, as well as the cascade of events that result in an adverse response relevant to a particular risk assessment question [15]. As with the fathead minnow database, the AOP approach leverages data in high-quality databases and existing information in the literature to build a weight of evidence to support the final assessment. In the process, AOP development helps in the identification of knowledge gaps, weaknesses, and strengths of association along the pathway. Efforts are currently under way to formulate procedures to help in the compilation, organization, and evaluation of the AOP WOE (e.g., alternate MOAs, how conserved the AOP is across taxonomic hierarchy, life-stage considerations, etc.), building on approaches developed for evaluating MOAs in human health risk assessments [16].

In 2007, The National Research Council identified the need for a paradigm shift in toxicity testing, one that reduces costs and the number of animals used, while increasing the number of chemicals tested through the use of biochemical and cell-based in vitro test assays [17]. To meet this goal, lessons can be learned from the development of the fathead minnow database. Despite the high-throughput nature of these new test protocols, with capabilities to deal with thousands of chemicals at a time, the results still need to be placed in a context that will help meet risk assessment goals. For instance, researchers still need to ensure that the chemical domain covers a diverse set of chemical spaces and mechanisms of action. The chemical space should also intersect sufficiently with concurrent high-quality, whole-organism data sets to allow for the development of reliable extrapolation models (e.g., in vitro to in vivo). Another issue is the availability and reliability of test assays. Under the Registration, Evaluation, Authorization, and Restriction of Chemicals (REACH) legislation it is estimated that for 72% of the chemical data gaps, acceptable alternative test methods are not available (e.g., in vitro or validated QSAR models) [18]. The assays used under this new testing strategy should include relevant molecular targets that are sufficiently sensitive systems for identifying the key events along the pathway. A final consideration is the long-term retention and public availability of study results and AOP information. High-throughput testing has the potential to generate many more results than observed in a traditional in vivo bioassay; therefore, it is necessary to plan for the long-term public storage of in vitro study results, and the development of tools to extract the data in a manner that is useful to risk assessors and researchers. The development of a comprehensive AOP database will be a critical research effort that will draw on even more diverse expertise than that used to establish the fathead minnow MOA knowledge base. Additional expertise in systems biology, bioinformatics, and omics research will be necessary. This new testing paradigm will require the availability of reliable in vitro data sets and robust computational models. Adherence to the basic principles used in the development of the fathead minnow database will ensure the long-term usefulness of these studies in establishing a more efficient and relevant risk assessment process.

SUPPLEMENTAL DATA

Table S1. (49 KB PDF).

Disclaimer—The views expressed in this work are those of the authors and do not necessarily reflect the views or policies of the US Environmental Protection Agency.

REFERENCES

1. Russom CL, Bradbury SP, Broderius SJ, Hammermeister DE, Drummond RA. 1997. Predicting modes of toxic action from chemical structure: Acute toxicity in the fathead minnow (*Pimephales promelas*). *Environ Toxicol Chem* 16:948–967.
2. Weininger D. 1988. SMILES, a chemical language and information system. 1. Introduction to methodology and encoding rules. *J Chem Inf Comput Sci* 28:31–36.
3. Russom CL. 2002. Mining environmental toxicology information: Web resources. *Toxicology* 173:75–88.
4. European Chemical Agency. 2009. Practical guide 5: How to report (Q) SARs. Report ECHA-10-B-10-EN. Helsinki, Finland.
5. Mayo-Bean K, Nabholz JV, Clements R, Zeeman M, Henry T, Rodier D, Moran K, Meylan B, Ranslow P. 2011. Methodology document for the ECOlogical Structure-Activity Relationship Model (ECOSAR) class program: Estimating toxicity of industrial chemicals to aquatic organisms using ECOSAR class program (Ver. 1.1). US Environmental Protection Agency, Office of Chemical Safety and Pollution Prevention, Office of Pollution Prevention and Toxics, Washington, DC.
6. Organization for Economic Cooperation and Development. 2007. Guidance document on the validation of (quantitative) structure-activity relationships (Q)SAR models. ENV/JM/MONO (2007) 2. Paris, France.
7. Veith GD, Greenwood B, Hunter RS, Niemi GJ, Regal RR. 1988. On the intrinsic dimensionality of chemical structure space. *Chemosphere* 17:1617–1630.
8. Bradbury SP, Russom CL, Ankley GT, Schultz TW, Walker JD. 2003. Overview of data and conceptual approaches for derivation of quantitative structure-activity relationships for ecotoxicological effects of organic chemicals. *Environ Toxicol Chem* 22:1789–1798.
9. Drummond RA, Russom CL. 1990. Behavioral toxicity syndromes: A promising tool for assessing toxicity mechanisms in juvenile fathead minnows. *Environ Toxicol Chem* 9:37–46.
10. McKim JM, Bradbury SP, Niemi GJ. 1987. Fish acute toxicity syndromes and their use in the QSAR approach to hazard assessment. *Environ Health Perspect* 71:171–186.
11. Broderius SJ, Kahl MD, Elonen GE, Hammermeister DE, Hoglund MDA. 2005. A comparison of the lethal and sublethal toxicity of organic chemical mixtures to the fathead minnow (*Pimephales promelas*). *Environ Toxicol Chem* 24:3117–2127.
12. Bradbury SP, Lipnick RL. 1990. Symposium on structural properties for determining mechanisms of toxic action. *Environ Health Perspect* 87:180–271.
13. Organization for Economic Cooperation and Development. 2004. OECD principles for the validation, for regulatory purposes, of (quantitative) structure-activity relationship models. The 37th Joint Meeting of the Chemicals Committee and Working Party on Chemicals, Pesticides and Biotechnology, Paris, France, November 17–19. Available from: http://www.oecd.org/dataoecd/33/37/37849783.pdf.
14. Bradbury SP, Feijtel TCJ, van Leeuwen CJ. 2004. Meeting the scientific needs of ecological risk assessment in a regulatory context. *Environ Sci Technol* 38:463A–470A.
15. Ankley GA, Bennett RE, Erickson RJ, Hoff DJ, Hornung MW, Johnson RD, Mount DR, Nichols JW, Russom CL, Schmieder PK, Serrrano JA, Tietge JE, Villeneuve DI. 2010. Adverse outcome pathways: A conceptual framework to support ecotoxicology research and risk assessment. *Enviro Toxicol Chem* 29:730–741.
16. Boobis SR, Doe JE, Heinrich-Hirsch B, Meek ME, Mun S, Rchirawat M, Schlatter J, Seed J, Vickers C. 2008. IPCS framework for analysing the relevance of a non-cancer mode of action for humans. *Crit Rev Toxicol* 38:87–96.
17. National Research Council. 2007. *Toxicity Testing in the 21st Century: A Vision and a Strategy*. National Academy of Sciences, Washington, DC.
18. van der Jagt S, Munn S, Torslov J, de Bruijn J. 2004. Alternative approaches can reduce the use of test animals under REACH. Report EUR 21405 EN. European Commission, Joint Research Center, Institute for Health Consumer Protection. Ispra, Italy.