CEFIC Long-range Research Initiative
Request for Proposals (RfP)

RfP (project code LRI-ECO20)

Title:
Development of an alternative testing strategy for the fish early life-stage (FELS) test (OECD 210)

Background

The use of conventional whole-organism (vertebrate) bioassays for estimating chronic ecological hazards has been limited due to low efficiency, high cost, extensive animal use, and, for aquatic toxicity studies, generation of large volumes of contaminated water. Moreover, most animal guideline tests provide little or no informative mechanistic toxicity data due to the principal focus on apical endpoints such as survival, growth, and reproduction. Due to these limitations, resource-efficient alternatives to conventional toxicity testing – including high-throughput *in vitro* and *in silico* screening assays – have been proposed as key components of a future testing paradigm for mechanism-based regulatory toxicology and ecotoxicology (Bradbury et al. 2004; NRC 2007; Villeneuve and Garcia-Reyero 2011). However, the predictive power of molecular or cellular perturbations (e.g., modeled or detected *in vitro*) for apical endpoints relevant to ecological risk assessment (i.e., survival, growth, and reproduction) must be sufficiently high to minimize uncertainties and provide meaningful data for regulatory decision-making.

To begin identifying useful predictive assays and testing strategies for regulatory ecotoxicology, Ankley and colleagues recently proposed the use of Adverse Outcome Pathways (AOPs) as a conceptual framework for summarizing existing knowledge about linkages between a direct, molecular-level initiating event and an adverse outcome at a level of biological organization relevant to ecological risk assessment (Ankley et al. 2010). Predictive linkages are implicitly defined when developing AOPs, providing a scientific framework for identification of targeted alternative assays and models that represent key events at multiple levels of biological levels of organization. Linked with biologically based, concentration-response models (e.g., toxicokinetic and toxicodynamic models), these alternative assays, once developed, can facilitate extrapolation to organism- and population-level endpoints and significantly decrease reliance on whole-animal regulatory toxicity tests as the primary source of hazard data for ecological risk assessment and environmental decision-making (NRC 2007).

The fish early life-stage (FELS) test (OECD 210) is the primary guideline used to estimate chronic toxicity of regulated chemicals (pesticides, industrial chemicals, pharmaceuticals, food/feed additives, and cosmetics). In addition, the FELS will be used to screen and prioritize hundreds to thousands of chemicals regulated under Registration, Evaluation, Authorization and Restriction of Chemical (REACH), Toxic Substances Control Act (TSCA), and other national or international chemical management programs. This effort will require development of a rapid, cost-efficient, and mechanism-based tiered testing strategy. For example, to begin addressing this challenge, development of a three-tiered testing strategy was recently proposed (Volz et al. 2011) as an outcome of an international meeting on development of approaches to address chronic toxicity and endocrine disruption testing needs in the context of animal alternatives. Volz et al. (2011) suggested an initial tier

representing high-throughput, adverse outcome pathway (AOP)-based *in vitro* (cell-based) assays (Tier 1); the second tier representing medium-to-high-throughput *in vivo* (zebrafish embryo) assays (Tier 2); and the highest tier representing the current FELS guideline test (Tier 3). In the long-term, implementation of a tiered testing strategy as a new paradigm for toxicity testing may help reduce reliance on long-term and costly FELS tests required for assessing the hazard of thousands of chemicals currently in commerce.

Objectives

The overall objective of this three-year research program is to initiate development of a tiered testing strategy for screening and prioritizing chemicals for FELS testing. Therefore, in order to accomplish the overall objective of this research program, the following objectives should be achieved by the end of the project period:

1) Identify, describe, and annotate toxicologically relevant FELS AOPs to provide a conceptual and scientific foundation for identification and development of Tier 1 assays.
2) Following annotation, identify and describe appropriate Tier 1 assays – as well as positive and negative reference chemicals – based on the catalog of FELS AOPs developed from Objective #1.
3) Develop and test Tier 1 and 2 assays using reference chemicals identified from Objective #2 and, based on comparisons of Tier 1 and 2 results, quantify false negative and positive error rates.
4) If data are not already available, test early life-stage toxicity of Tier 1 and 2 reference chemicals using a modified OECD 210 guideline that includes endpoints reflecting AOPs represented within the Tier 1 assays and, based on these results, quantify the ability of Tier 1 and 2 assays to predict fish early life-stage toxicity.

Scope

The expected outcome of this three-year project is to provide the first critical steps for development of an alternative testing strategy for a vertebrate bioassay (OECD 210) widely used to predict chronic fish toxicity and support ecological risk assessments around the world. Given that the OECD 210 is an internationally used harmonized guideline, investigators are expected to develop plans for seeking input and guidance from key colleagues and scientists within academia, industry, and governmental agencies around the world during the project period to ensure that this strategy will be successful and valuable for the global scientific and regulated community.

LRI Funding

€500,000

Timing

Project to start beginning 2013 for up to 3 years

Partnering/Co-funding

Applicants should provide an indication of additional partners and funding opportunities that can be appropriately leveraged as part of their proposal. Partners can include, but are not limited to industry, government/regulatory organizations (e.g. US EPA, OECD,…), research institutes, etc. Statements from potential partners should be included in the proposal package.

References

Ankley GT, Bennett RS, Erickson RJ, Hoff DJ, Hornung MW, Johnson RD, et al. 2010. Adverse outcome pathways: A conceptual framework to support ecotoxicology research and risk assessment. Environ Toxicol Chem 29(3): 730-741.

Bradbury SP, Feijtel TC, Van Leeuwen CJ. 2004. Meeting the scientific needs of ecological risk assessment in a regulatory context. Environ Sci Technol 38(23): 463A-470A.

NRC. 2007. Toxicity testing in the 21st century : a vision and a strategy. Washington, DC: National Academies Press.

Villeneuve DL, Garcia-Reyero N. 2011. Vision & strategy: Predictive ecotoxicology in the 21st century. Environ Toxicol Chem 30(1): 1-8.

Volz DC, Belanger S, Embry M, Padilla S, Sanderson H, Schirmer K, et al. 2011. Adverse outcome pathways during early fish development: a conceptual framework for identification of chemical screening and prioritization strategies. Toxicol Sci 123(2): 349-358.

DEADLINE FOR SUBMISSIONS: August 31, 2012
Please see www.cefic-lri.org for the project proposal form and further guidance for grant applications.