ICCVAM Communities of Practice Webinar – January 27, 2015

Reverse Toxicokinetics: Using In Vitro Data to Estimate Exposures that Could Be Associated with Adverse Effects In Vivo

Additional information and materials available at http://ntp.niehs.nih.gov/go/ivive-webinar

Overview
High throughput screening (HTS) assays, such as those used in Tox21 and ToxCast, provide promise for rapidly screening chemicals for potential toxicological effects. However, differences in bioavailability and clearance between in vitro and in vivo systems make it difficult to directly correlate effective test chemical concentration in an in vitro assay with the in vivo dose that could cause toxic effects. Extrapolation from in vitro to in vivo results must account for these differences and consider which toxicokinetic (TK) factors are most relevant. Dr. John Wambaugh, from the EPA National Center for Computational Toxicology, and Dr. Barbara Wetmore, from the Hamner Institutes of Health Sciences provided examples of how these concepts can be applied.

Summary of Dr. Wambaugh’s Presentation
Dr. Wambaugh began the webinar by giving an overview of how to develop reverse TK (RTK) models to correlate in vitro and in vivo activity. RTK is used to convert effective in vitro HTS results to daily doses that produce similar serum levels in humans for comparison to exposure data. Paramount to building an RTK model is the assessment of in vitro bioactivity. Since most HTS assays in Tox21 and ToxCast were conducted in a dose–response format, a pre-defined “active” concentration can be selected as the bioactivity metric (e.g., the concentration at 50% maximal activity [AC50]). Dr. Wambaugh briefly described the high throughput TK (HTTK) assays (i.e., in vitro plasma protein binding and metabolic clearance) that allow approximate hepatic and renal clearances to be calculated. Data from these assays can be integrated with in vitro bioactivity to generate an estimate of the steady-state concentration (Css) and the dose required to induce such an effect in vivo.

This approach is borrowed from the pharmaceutical industry, which has applied these models for years to estimate efficacious doses and the success of planned clinical trials. Ultimately, the results of such an approach can be directly compared to clinical data. However, for environmental compounds human data are often not available and therefore the uncertainty of these estimates must be characterized. Where data are available, this can be done directly by calculating the discrepancy between the in vitro Css predicted from HTTK and in vivo Css values determined from the literature (i.e., the residual).
Computer models can also be used to predict when the residual will be small based on chemical
descriptors (physicochemical properties, in vitro HTTK data, QSAR to predict chemicals that are
substrates for active transport), and hence indicate when RTK will be most accurate. A recursive
partitioning tree is one approach that can be used to predict the residual, which can then be used as a
central tenet was to
gerah chemica-specific estimate of the accuracy of HTTK predictions. Dr. Wambaugh also described applying
more complex physiologically based TK models that incorporate more realistic, sporadic dosing
simulations to test some of the assumptions inherent to RTK models (constant, uniform infusion;
sufficient time interval to reach steady state). Regardless of the approach, the central tenet was to
carefully characterize uncertainty in any approach used for IVIVE.

Dr. Wambaugh closed by looking to future efforts to further improve evaluation data and thereby allow
better assessment of predictive ability and determination of domain of applicability including:

- Improved approaches to collecting HTTK data
- Expanding population variability beyond just adults
- Collecting data from limited in vivo studies (EPA/NHEERL and Research Triangle Institute)
- Organizing data from larger, systematic studies (e.g., National Toxicology Program) into
  computable form

Summary of Dr. Wetmore's Presentation

Dr. Wetmore followed with a presentation of a comprehensive attempt to combine physiologic and
pharmacokinetic (PK) differences to quantitate variability anticipated between age, ethnic, and disease-
based populations.

Incorporating population variability and sensitive subpopulations into dosimetry for high-throughput
toxicity testing is an important consideration to more accurately perform IVIVE. Dr. Wetmore
highlighted recent efforts to better inform risk assessment by integrating dosimetry and exposure with
HTS data, but noted that such a strategy is limited to the general population and does not take into
account susceptible subpopulations (e.g., juvenile or geriatric). Reliance on PK data for a “generic”
population could lead to a significant underestimation of risk to a susceptible subpopulation. In an attempt
to account for this inherent variability, population based PK modeling and simulation tools can be used to
predict outcomes in relevant patient populations rather than a single value representing an average
individual.

Dr. Wetmore also described how recombinant phase I and II enzyme assays contribute to population-
based IVIVE extrapolations. Incorporating recombinant enzyme metabolism and human plasma protein
binding data in an IVIVE model allows for steady-state plasma concentrations derived and compared
across multiple subpopulations (e.g., ethnicity, age, disease state). Chemical-specific adjustment factors can then be estimated and compared to oral equivalent doses and exposure estimates for subpopulations. This approach defines a range of PK variability when comparing the general population to the most sensitive population. These subpopulation-based differences will also contribute to the variable susceptibilities in the entire population that may be observed following chemical exposure. This approach demonstrates the feasibility of measuring isozyme-specific clearance rates and using them to capture population variability for industrial chemicals.

Clearly, identifying and accounting for uncertainty and variability are paramount to an optimal IVIVE approach. Drs. Wambaugh and Wetmore highlighted efforts underway to ensure that IVIVE using ToxCast and Tox21 data are addressing these considerations.