317.2 hr, which differ by 72.6 hr (3 days). We used stochastic computational modeling to clarify the biological underpinnings of these different values of D. The value of D depends on the growth kinetics of the tumor cells. Stochastic simulation used pseudorandom numbers from binomial and Poisson distributions to predict the division and death of tumor cells. The probability of division per hour was set to an upper bound on the rate of cell division (0.021) in rats exposed chronically to 9.93 ppm formaldehyde by inhalation. Cell volume (3.4e-9 cm3) and the minimum size of clinically detectable tumors (0.1 to 1.0 cm3) were estimated from relevant literature. With these parameters fixed, the probability of cell death was varied to achieve different values of D. Some tumors became extinct, even when the probability of division was greater than the probability of death. Average values for D and its upper and lower bounds were obtained by running the model repeatedly, typically from 1,000 to 10,000 times. These analyses showed that the values of D used by Subramaniam et al. (2007) are associated with little or no cell death as the tumor grows and that the 3day difference in D between incidental and fatal tumors would probably not be identifiable, given the stochastic variability of the system. The value of D used by Conolly et al. (2003) was associated with a relatively large death rate, with many of the tumors becoming extinct, and is consistent with the cytotoxic environment created by ongoing exposure to 9.93 ppm formaldehyde. Furthermore, p53 mutation, which is associated with inactivation of the apoptotic pathway, is not a consistent feature of formaldehyde-induced tumors in the rat nose. This genomic observation is consistent with a larger dearth rate and with the larger value of D identified by Conolly et al. (2003). This analysis increases confidence in the value of D used by Conolly et al. (2003) and in their BBDR modeling in support of cancer risk assessment for formaldehyde.

3827 Moving beyond Similarity Scores: Use of Toxicokinetic and Toxicodynamic Data to Inform Selection of a Suitable Analogue for δ-HCH in Read-Across Assessment

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Delta hexachlorocyclohexane (δHCH) has limited toxicological data that are not adequate for traditional derivation of toxicity values. Instead, a tiered analogue approach relying on assessment of structural, metabolic, and toxicological similarity between the target chemical and potential analogues was explored. This read-across case study highlights the utility of looking beyond similarity scores and examining all available data in selecting an analogue. Structural analogues identified for δ-HCH included three of its stereoisomers (α-, β-, and γ-HCH). Critical endpoints identified from subchronic and chronic oral studies of these stereoisomers were liver and immune system effects. The stereoisomers each score 100% for structural similarity with the target compound (δ-HCH) and share the same structural alerts and molecular weight as the target. However, toxicokinetic data showed that β-HCH undergoes very little metabolism in mammals and has a much longer half-life in the body than δ-, α, or γHCH. Furthermore, comparative toxicity data on the hepatotoxicity of the HCH stereoisomers revealed similarities between δ-HCH and γ-HCH and differences from the other two stereoisomers. Ito et al. (1973 and 1975) examined liver effects in mice and rats, respectively, after 24-to-48-week administration of δ, α, β , and γHCH. In these studies, dose-response relationships for liver weight and histopathology changes were similar between δ-HCH and γ-HCH, while α- and βHCH showed greater potency (larger magnitudes of effect at lower doses) for hepatotoxic effects in both species. *In vitro* data also suggested that, of the HCH stereoisomers, δHCH is most similar to γHCH with respect to immune system effects. Both δ- and γ-HCH inhibited mitogenic response to phytohemagglutinin in bovine lymphocytes while α- and βHCH did not. Based on structural similarity, physicochemical properties, toxicokinetics, and toxicodynamic comparisons for sensitive endpoints including liver toxicity and immune system effects, γHCH was selected as the most suitable analogue for δHCH. This case study demonstrates how toxicokinetic and toxicodynamic information can be used to refine selection of analogues in read-across assessment. *The views expressed are those of the authors and do not necessarily reflect the views and policies of the US EPA.*

3828 A Systematic Comparison of the Temporal Transcriptional Responses by Hepatotoxicants in Primary Human Hepatocytes and HepaRG Cells Using Concentration Response Modeling of Gene Co-expression Networks

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Next generation risk assessment (NGRA) of chemicals revolves around the use of mechanistic information without animal experimentation. In this regard, toxicogenomics has proven to be a useful tool to elucidate underlying mechanisms of toxicological action of adverse effects of xenobiotics. In the present study, two widely used human hepatocyte culture systems, namely primary human hepatocytes (PHH) and human hepatoma HepaRG cells, were exposed to chemical liver toxicants known to induce liver steatosis, cholestasis or necrosis. Benchmark dose (BMD) response modelling was applied to transcriptomics gene co-expression networks (modules) in order to derive benchmark concentrations (BMCs) and to gain mechanistic insight into the hepatotoxic effects. BMCs derived by benchmark dose modelling of gene co-expression modules recapitulated benchmark dose modelling of individual genes. PHH and HepaRG cells showed overlap in the deregulated genes and modules by the liver toxicants. However, PHH demonstrated more biological activity compared to HepaRG cells, based on the lower BMCs of co-regulated gene modules, which could be used as point of departure for the associated cellular (stress) pathways/processes. This approach could serve next generation risk assessment practice to identify early responsive modules at low benchmark concentrations. In turn, this can assist in delineating potential hazards of new test chemicals using *in vitro* systems. Benchmark concentrations may be paired with chemical exposure assessment and used in a subsequent risk assessment. *This project has received funding from Cosmetics Europe and the European Chemical Industry Council (CEFIC) (project AIMT10), the EC Horizon2020 EUToxRisk project (grant number 681002), the EC Horizon2020 RISK-HUNT3R project (grant number 964537; part of ASPIS cluster) and the EU-EFPIA Innovative Medicines Initiative 2 (IMI2) Joint Undertaking under the TransQST project (grant number 116030). This Joint Undertaking receives support from the European Union's Horizon 2020 research and innovation program and EFPIA.*

3829 Systematic Application of Mode-of-Action and Human Relevance Analysis: Styrene-Induced Lung Tumors in Mice

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Risk assessment of human health hazards often relies on observations from animal experiments. Although exposure studies in rats and mice are a major basis for determining risk in many cases, observations made in animals do not always reflect health hazards in humans due to differences in biology. In this critical review, the mode-of-action (MOA) human relevance framework was used to assess the likelihood that bronchiolar lung tumors observed in mice chronically exposed to styrene represent a plausible tumor risk in humans. Available datasets were used to determine the weight-of-evidence 1) that styrene-induced tumors in mice occur through a MOA initiated by metabolism of styrene by Cyp2F2; and 2) whether the hypothesized key event relationships are plausible in other species. Analysis of data using five modified Hill causality considerations indicated that the hypothesized Cyp2F2-dependent MOA is active in mice, but only results in tumorigenicity in susceptible strains. Assessment of species concordance to determine whether analogous key event relationships do or could plausibly occur in other species concluded that while some of the proposed key events are biologically plausible in rats, the MOA is improbable in humans due to poor concordance of both early and late key events reflecting key differences in airway biology and physiology. This analysis serves as a rigorous demonstration of the framework's utility in increasing transparency and consistency in evidence-based assessment of MOA hypotheses in toxicological models and determining relevance to human health.

3830 Interindividual Variability Assessments through Benchmark Dose-Response Modeling of Primary Human Bronchial Epithelial-Fibroblast Co-culture Responses to Acrolein

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Human health risks are known to significantly vary according to individuals across populations; however, this interindividual variability cannot be captured by traditional *in vivo* or *in vitro* studies that utilize inbred animal strains and isogenic cell lines, respectively. The use of *in vitro* systems that utilize cells isolated directly from a range of individual human donors (*i.e.*, "primary" cells) provides a novel opportunity to simultaneously increase the physiological and public health relevance of *in vitro* new approach methodologies. Unfortunately, the utility of incorporating interindividual variability into *in vitro* testing has not yet been integrated into toxicity testing strategies due to a lack of computational methods and software infrastructure. To address this gap, we hypothesized that benchmark dose-response (BMD) modeling could be leveraged to evaluate human interindividual sensitivities to chemical exposures. Here, we performed BMD modeling on a dataset containing a broad range of endpoints reflective of *in vivo* tissue physiology. Endpoints were derived from an inhalation assay battery, including 6 phenotypic and 11 secreted cytokine/growth factor endpoints, to evaluate the effect of acute exposure to the ubiquitous reactive volatile organic gas acrolein (0-4 ppm) on primary human bronchial epithelial-fibroblast co-cultures (n=14). We then clustered observations using model curve fit parameters to assess whether dose-response was associated with cell donor demographic variables. This study identified the following: First, we found that benchmark doses varied greatly on a per-donor basis, with BMDs spreading up to 10-fold after model averaging approaches (e.g., BMDs for vascular endothelial growth factor A ranged 0.84-7.32 ppm [at benchmark responses of 2 SD]). Second, BMDs were generally lower when analyzing response trends on a per-donor basis, as opposed to aggregating averaged responses across donors; this demonstrated the need to capture and quantify sensitive populations by evaluating individual-specific

62nd Annual Meeting & ToxExpo
Nashville, TN • March 19–23, 2023

The Toxicologist

Supplement to *Toxicological Sciences*

SOT | Society of Toxicology

Toxicological Sciences

The Official Journal of the Society of Toxicology

OXFORD UNIVERSITY PRESS

ISSN 1096-6080 Volume 192, Issue S1 March 2023
www.academic.oup.com/toxsci

Publication Date: March 14, 2023