Predicting Developmental Toxicity of ToxCast Phase I Chemicals Using Human Embryonic Stem Cells and Metabolomics

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EPA's ToxRefDB contains prenatal guideline study data from rats and rabbits for over 240 chemicals that overlap with the ToxCast *in vitro* high throughput screening project. A subset of these compounds were tested in Stemina Biomarker Discovery's developmental toxicity platform, an *in vitro* method combining human embryonic stem (hES) cells and metabolomics to discover biomarkers of developmental toxicity. The purpose of this pilot study was to perform LC-MS based nontargeted metabolomic analysis on the supernatant of hES cell cultures dosed with blinded EPA test chemicals to identify human metabolites subject to chemically induced alterations and biochemical signatures which may be indicative of potential human developmental toxicity. Significant fold changes in endogenous human metabolites were detected for 83 annotated mass features in response to the subset of ToxCast chemicals. The annotations were mapped to specific human metabolic pathways with nicotinate and nicotinamide metabolism, pantothenate and CoA biosynthesis, glutathione metabolism, and arginine and proline metabolism pathways most affected. Stemina's predictive DevTox® model, trained on 22 pharmaceutical agents of known teratogenicity and differing potency, was applied to the blinded EPA test compound data to test predictivity for mammalian *in vivo* data. The model correctly predicted teratogenicity for eight of eleven compounds and an additional compound showed agreement with animal data at the low treatment dose. Thus, our initial results indicate this platform as an alternative to animal models for predicting developmental toxicity and providing mechanistic information about the underlying biochemical pathways. [This abstract does not necessarily reflect US EPA policy]