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IN VITRO SYSTEMS

A transcriptome-based classifier to identify developmental toxicants by stem cell testing: design, validation and optimization for histone deacetylase inhibitors

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Abstract Test systems to identify developmental toxicants are urgently needed. A combination of human stem cell technology and transcriptome analysis was to provide a proof of concept that toxicants with a related mode of action can be identified and grouped for read-across. We chose a test system of developmental toxicity, related to the generation of neuroectoderm from pluripotent stem cells (UKN1), and exposed cells for 6 days to the histone deacetylase inhibitors (HDACi) valproic acid, trichostatin A, vorinostat, belinostat, panobinostat and entinostat. To provide insight into their toxic action, we identified HDACi consensus genes, assigned them to superordinate biological processes and mapped them to a human transcription factor network constructed from hundreds of transcriptome data sets. We also tested a heterogeneous group of 'mercurials' (methylmercury, thimerosal, mercury(II)chloride, mercury(II)bromide, 4-chloromercuribenzoic acid, phenylmercuric acid). Microarray data were compared at the highest non-cytotoxic concentration for all 12 toxicants. A support vector machine (SVM)-based classifier predicted all HDACi correctly. For validation, the classifier was applied to legacy data sets of HDACi, and for each exposure situation, the SVM predictions correlated with the developmental toxicity. Finally, optimization of the classifier based on 100 probe sets showed that eight genes (F2RL2, TFAP2B, EDNRA, FOXD3, SIX3, MT1E, ETS1 and LHX2) are sufficient to separate HDACi from mercurials. Our data demonstrate how human stem cells and transcriptome analysis can be combined for mechanistic grouping and prediction of toxicants. Extension of this concept to mechanisms beyond HDACi would allow prediction of human developmental toxicity hazard of unknown compounds with the UKN1 test system.

Eugen Rempel and Lisa Hoelting shared first authorship.

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