Development and Validation of a Computational Model for Androgen Receptor Activity

N Kleinstreuer[1], P Ceger[2], D Allen[2], P Browne[3], K Houck[4], M Martin[4], R Thomas[4], D Dix[3], W Casey[1], R Judson[4]

[1]NIH/NIEHS/DNTP/NICEATM, RTP, NC, USA, [2]ILS, RTP, NC, USA; [3]EPA/OCSPP/OSCP, Washington, DC, USA; [4]EPA/ORD/NCCT, RTP, NC, USA

Screening thousands of chemicals to identify androgen receptor (AR) agonists or antagonists would require millions of dollars, thousands of animals, and decades of time using current OECD or U.S. EPA validated methods. Alternative methods developed in the Tox21 and ToxCast research programs incorporate High-Throughput Screening (HTS) assays and computational pathway models to rapidly and inexpensively identify potential endocrine-active chemicals. Eleven Tox21/ToxCast HTS AR assays were integrated in a computational model of the AR pathway to distinguish true activity from technology-specific assay interference. The HTS assays assess potential activity at multiple points of the AR pathway (receptor binding, cofactor recruitment, gene transcription and protein production) in multiple cell types. The Tox21 antagonist transactivation assay in MDA-kb2 cells was run with different concentrations of the synthetic AR ligand R-1881 to confirm activity specific to AR antagonism. These confirmation data were combined with cytotoxicity data from multiple assays to provide further insight into potential non-specific activity and provide a confidence score for true AR pathway activity. Validation of results from such alternative screening methods requires a robust set of reference chemicals; therefore we compiled data on 158 putative androgen-active or inactive reference chemicals from international AR test method validation efforts. We conducted semi-automated literature reviews for in vitro AR binding and transactivation assays on these chemicals, and extracted detailed assay information and results from identified references into a single database using a standardized ontology. Based on quantitative data such as activating (or inhibiting) concentrations, we identified reference chemicals with consistent results and assigned potency ranges. The AR pathway model based on Tox21/ToxCast data predicted AR activity with 93% (27/29) and 97% (27/28) accuracy for agonist and antagonist reference chemicals, respectively. The model was used to screen 1853 chemicals, identifying 173 as AR agonists or antagonists. An additional 274 chemicals were predicted to have very weak AR pathway activity. The Tox21/ToxCast HTS data support a biologically based computational model distinguishing assay interference from true AR pathway activity and rapidly screening large numbers of environmental chemicals for androgenic or anti-androgenic activity. *This work does not reflect the official policy of any federal agency.*

Presenting author: Nicole Kleinstreuer, Nicole.kleinstreuer@nih.gov