In Vitro Screening of 1877 Industrial and Consumer Chemicals, Pesticides and Pharmaceuticals in up to 782 Assays: ToxCast Phase I and II

R Judson, K Houck, M Martin, A Richard, T Knudsen, N Sipes, I Shah, S Little, J Wambaugh, W Setzer, J Rabinowitz, M Linnenbrink, J Franzosa, P Kothiya, J Phuong, K McLaurin, D Filer, MCK Leung, C Strope, L Truong, R Thomas, D Smith, D Reif, D Rotroff, N Kleinstreuer, M Xia, R Huang

In Phase II of the ToxCast program, the U.S. EPA and Tox21 partners screened 1,877 chemicals, including pesticides; food, cosmetics and personal care ingredients; pharmaceuticals; and industrial chemicals. Testing used a 782 in vitro assays across 7 technologies and multiple biological formats (cell-free, cell lines and primary cells from multiple tissues). Assays were run in concentration-response format (from ~0.01 to ~100 μM) with replicates and controls. We report several key findings. First, in 91% of the genes assayed for which we have reference chemicals (defined as a chemical with a known molecular target, 75 out of 313 genes total), a reference chemical is in the top quartile of potency. Second, 41% of chemicals show a "burst" of non-specific activity related to cell stress / cytotoxicity. On average, once ~5% of assays are activated by a chemical, there is a linear increase in the number of cytotoxicity/cell-stress-related hits relative to hits in other assays (R2=0.84). This indicates a potential for assay activity which is not due to the chemical interacting with the intended assay target. In a set of 40 chemicals with both ToxCast data and in vitro pharmacokinetic data, the burst concentration corresponds roughly to concentrations predicted to be seen in vivo at the maximum tolerated dose (MTD) in rat 2 year chronic cancer studies (78% of MTDs were in the burst region). Potent cholinesterase inhibitors were a prominent exception, with MTDs all below the burst range, since the MTD is driven by cholinesterase activity rather than systemic effects. Finally, chemicals were ranked by relative potency and target specificity to provide a quantitative metric to prioritize chemicals for further study. About ~1% of chemical-gene combinations show potent and specific activity, defined as AC50<10□M and below the burst, which is ~3 hits per chemical on average. This abstract does not necessarily reflect Agency policy.