

## High Throughput Screening (HTS) Assays Workshop

The NTP held a workshop on December 14 and 15, 2005, at the Hyatt Crystal City, Arlington VA. This workshop was an NTP Roadmap activity to receive input on HTS technology and the approach the NTP should take to implement these assays in its testing program. Approximately 70 attendees representing academia, industry, U.S. Federal agencies, external NTP advisory groups, and animal welfare groups participated.

### *1. Objectives*

The workshop's objectives were to:

- (1) Educate the NTP on how HTS assays are typically conducted.
- (2) Identify which HTS assays might be the most informative in terms of possibly predicting toxicological responses in laboratory animals and humans.
- (3) Recommend how chemicals might be selected/prioritized for testing in HTS assays.
- (4) Describe how the HTS assay data should be collected, stored, and mined.
- (5) Consider how U.S. Federal agencies could use information from these assays in making regulatory decisions.

### *2. Format*

The meeting began with a plenary session (agenda attached) at which seven presentations were made. During this session the interaction between the Molecular Libraries Initiative (MLI), which is part of the NIH Strategic Plan, and the NTP's HTS initiative was discussed. Following the plenary session, the attendees were divided among four breakout groups to address specific questions related to targets and assays for HTS; study design, chemical selection, and analytical methods; data storage, analysis, and interpretation; and application of data from HTS assays to regulatory decision-making. The attendees reconvened mid-morning in plenary session on the second day to discuss the deliberations of the individual breakout groups.

Findings from the workshop will be presented to the NTP Board of Scientific Counselors at its meeting on June 13, 2006. The following text summarizes the recommendations of the breakout groups at the workshop and outlines NTP's next steps with regard to the HTS initiative.

### *3. Workshop Recommendations*

#### a. Targets and Assays for HTS

The breakout group used a holistic approach to identify pathways and targets, and recommended that studies include multi-dose and multi-time points and include testing on primary rodent and human cells with metabolizing capacity. The group focused on assays and pathways relevant to carcinogenesis and identified six critical pathways namely, apoptosis, proliferation and cell cycle control, DNA damage and repair, chromatin remodeling, modulation of signal transduction, and the p53 pathway. The group then identified cell types and possible endpoints to measure for each of the pathways.

#### b. Study Design, Chemical Selection and Analytical Methods

The breakout group outlined the ideal experimental approach for an HTS study realizing that this might not be achievable. They suggested (1) that a dose-response curve be developed for each chemical tested, (2) that each study be replicated at least once, (3) that standards and concurrent positive and negative controls be run daily, and (4) that a historical reference chemical be included in each study. The group recommended that the NTP prioritize chemicals for testing based on public concern, and that the NTP also test any metabolites of the chemicals, if known and available. They recommended that the NTP construct a database with software for chemoinformatics to house the HTS data, and hire experienced personnel to compile and manage the database.

#### c. Data Storage, Analysis and Interpretation

The breakout group recommended that the data be stored in its raw format and have appropriate structural annotations for identification. With regard to analysis of the HTS data, the group suggested that the data be normalized to the internal controls used in the specific study. The group supported making the HTS data accessible to the public, easily retrievable from a database, and linked to the NTP databases containing toxicological information on these chemicals. The NTP noted that the data on NTP chemicals generated by the MLI would be stored in the PubChem database and recommended that all data collected in the future from the NTP should also be stored in this database. They recommended also that the HTS database be constructed to enable easy retrieval of data for chemicals with similar structures and that it be linked with the current NTP database to retrieve information on toxicological responses.

Technical details for this access would entail setting up essential handshaking steps between the two databases and translating the Chemical Abstracts Registry Numbers to machine language so that bioassay data could be retrieved along with the HTS data.

#### d. Application of Data from HTS Assays in Regulatory Decision-making

The breakout group agreed that HTS assay data could not be used presently for regulatory decision-making. However, the group thought that the data would be useful to help set priorities for additional toxicological testing and that the assays would help to reduce, refine, and replace animal testing. They noted a need for workshops to train personnel in regulatory agencies on the use and proper interpretation of HTS data. The group pointed out that acceptance of data from HTS assays by the scientific community, U.S. regulatory agencies, and the public would require demonstration of relevance and reliability. The group recommended that the NTP include education and communication strategies in its HTS initiative.