During the past few years, scientists have developed new technologies that can improve our understanding of biological responses to chemicals in human tissues. Genomics (the study of genes), proteomics (the study of proteins), and other technologies offer a promising solution for evaluating the effects of environmental chemicals. Genomics, for example, will allow researchers to identify changes in the expression (activation or repression) of individual genes after exposure to chemicals, and potentially link these changes to specific biological responses.

Much of the promise of these new technologies stems from their capacity to simultaneously measure changes in thousands of genes or proteins and their ability to do this rapidly for many different chemicals. This wealth of information provides a much broader picture of the potential biological effects of a chemical than the current animal toxicological bioassays. However, interpreting the enormous amount of data that results from these new technologies and applying the results to risk assessment is a daunting task (NRC, 2007a; NRC, 2007b). For example, a single microarray test of gene expression changes following several doses of one chemical may result in over one million data points. A rigorous statistical tool is essential to link this barrage of data to changes in the body and to a defined increase in risk.

A research project led by Dr. Rusty Thomas of The Hamner Institutes for Health Sciences, co-sponsored by the American Chemistry Council’s Long-Range Research Initiative and the Formaldehyde Council, Inc., focused on integrating a common risk assessment method, known as “benchmark dose” (BMD), with gene expression microarray data to assess the effects of graded doses of chemical exposure. By integrating the BMD method with gene expression microarray data, the researchers performed a comprehensive survey of molecular and cellular changes associated with chemical exposure and identified doses at which different cellular processes are altered. Because the effects on individual genes can be measured at low doses, they can be compared with real-world exposures to estimate whether humans might be at risk.

Making Sense of Genomic Data: a Dose-Response Analysis Approach

“We have created a new software tool that helps interpret the enormous amounts of genomic data in a risk assessment context, allowing us to link cellular/tissue alterations with specific doses.”

—Dr. Rusty Thomas, The Hamner Institutes for Health Sciences

BMDExpress: A New Tool to Assess the Health Effects of Chemical Exposure

A novel software tool, BMDExpress, was developed to enable interpretation of the tens of thousands of data points arising from commonly used genomic assays. The program creatively combined the following three current techniques:

- **Microarray analysis**, which can rapidly measure dose-response changes in thousands of genes following chemical exposure.
- **Benchmark dose analysis**, which is a common risk assessment method to calculate the “benchmark” dose producing minimal or no adverse health effects. This dose is then used by regulatory agencies to set safe exposure levels.
- **Gene function (also called gene ontology) analysis**, which is used to group genes based on their known biological functions and evaluate whether changes in specific genes identified using microarrays correspond to effects on important biological functions, such as the immune system, cell death, etc.

Background

**Genetic Signatures of Biological Effects**

**Response**

BMD

Dose

Approach

Microarray Test

Benchmark Dose

Genes A-D=immune response

Genes E-M=cell death

Genetic Signatures of Biological Effects

**BMDExpress**

The Long-Range Research Initiative (LRI), a program of the American Chemistry Council, sponsors research that increases scientific knowledge of the potential impacts that chemicals may have on human health, wildlife, and the environment. Results are publicly available. This LRI Perspective is one in a series of documents that summarize LRI-sponsored research. See www.americanchemistry.com/lri.
The researchers developed a unique risk assessment software tool, called BMDExpress, to analyze dose-response relationships on a molecular level. The tool combines BMD calculations with a special type of gene function analysis (e.g., identifying whether specific genes responsible for a biological function, such as lung cellular integrity, are altered) in the processing of gene expression microarray data. The combination of microarray technology and the BMDExpress software tool allows a comprehensive survey of molecular changes following chemical exposure, as well as dose estimates at which different cellular processes are altered.

To test BMDExpress, the researchers conducted two case studies. The first looked at short-term, inhalation exposure of rats to formaldehyde to evaluate dose-response changes in nasal tissue and identify how the chemical may cause tumors. The second case study examined genomic changes in zebrafish that were exposed to different doses of an endocrine-active compound (an estrogen used in birth control pills) to identify sets of genes that may be potential biomarkers of such compounds in fish.

The results demonstrate that BMDExpress can integrate genomic information directly into the risk assessment process. In the first case study, researchers performed gene expression microarray analysis on cells isolated from the noses of rats exposed for brief periods to formaldehyde, and then applied BMDExpress to evaluate dose-response relationships for all the different genes. The results compared well with previous studies of rats similarly exposed for longer periods of time. Importantly, the genomic changes were no more sensitive than traditional toxicological methods that detected tumors and other cellular changes, but provided a less biased and more broad-based way to survey potential perturbations in the wide range of biological processes that are important in a tissue. The BMDExpress software also identified doses at which these perturbations occurred and helped to understand the potential mechanism underlying these perturbations.

In the second case study, comparisons of results from BMDExpress and traditional methods of gene expression microarray analysis on fish exposed to estrogenic compounds highlighted the software tool’s utility for identifying potential biomarkers for ecotoxicology.

The results from this work represent a significant step forward in rigorously applying genomic information to the risk assessment process. The new risk assessment guidelines by the Environmental Protection Agency allow for evaluations based on mode-of-action, which is a general understanding of how a chemical causes a specific toxic response. The dose-response analysis of genomic data performed by BMDExpress may identify changes in specific biological processes that could be used in a mode-of-action risk assessment. This would help define the chemical’s behavior in the low-dose region instead of relying on the traditional default values that are based heavily on assumptions. To facilitate its broader use both nationally and internationally, this BMDExpress software is now freely available to all interested parties (http://sourceforge.net/projects/bmdexpress/).

References


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