Towards an Innovative DNA Array Technology for Detection of Pharmaceuticals in Reclaimed Water

Numerous pharmaceutical substances and their metabolites have been identified in the aquatic environment. The general premise of this research is that pharmaceuticals, at environmentally relevant concentrations, will elicit molecular changes in exposed organisms that can be quantified and employed as a bioanalytical tool for exposure assessment.

This research investigated the use of DNA microarrays as a bioanalytical tool for analysis of pharmaceuticals in treated waters that are intended for reuse.

This project developed a bioassay that targets molecular events associated with pharmaceutically induced gene induction and repression in exposed surrogate organisms. The objective is to use pharmaceutically induced gene changes as a means to assess exposure to—and thus their presence in—aquatic environments. This bioassay approach permits evaluation of biological effects of pharmaceuticals and significantly enhances ongoing efforts to understand and address the short- and long-term consequences of these compounds in the environment.

Recent interest in quantifying pharmaceuticals in waste effluents and aquatic environments has highlighted the need for bioanalytical assays that will accurately reflect the presence of these compounds. This research investigated the use of DNA microarrays as a bioanalytical tool for analysis of pharmaceuticals in treated waters that are intended for subsequent further use, i.e. reuse waters.

Background

U.S. surface waters are subjected to increasing anthropogenic stress, coupled with natural stressors such as salinity, dissolved oxygen, temperature, nutrient inputs, and physical energy. River and coastal waters/sediments are the ultimate recipients of an array of pollutants released in municipal and industrial point discharges, and carried in urban and agricultural non-point runoff. River and coastal regions throughout the U.S. are impacted by chemical stressors including pesticides, polycyclic aromatic hydrocarbons (PAHs) and polychlorinated biphenyls (PCBs) and pharmaceuticals and personal care products (PPCPs).

With unique chemical and physical characteristics, analytical procedures for analysis of pharmaceuticals in water matrices have proven difficult. This is due in part to the large numbers of compounds, the low concentrations in which they are found in the environment, and inability to develop broad extraction procedures. To circumvent these issues, biological methods are becoming increasingly popular as broad-based environmental screening tools. Successful bioassays have been designed to screen for some pharmaceuticals such as estrogenic or androgenic environmental endocrine disruptors (EDCs), but these represent a small subset of pharmaceuticals and, to date, bioassays for most pharmaceutical compounds have not been developed.

BENEFITS

- Identifies new environmental biomarkers by analysis of induction and/or repression of select gene markers and recognition of specific gene expression signatures.
- Establishes validity of using DNA arrays to rapidly screen changes in gene expression in response to aquatic pharmaceutical exposure.
- Improves environmental monitoring and accelerates the ability to rapidly assess chemical exposure.
- Simultaneously screens thousands of genes eliminating the time and cost of traditional biomarker analysis.

RELATED PRODUCTS

Online Methods for Evaluating the Safety of Reclaimed Water (01HHE4a)

Removal of Endocrine Disrupting Compounds in Water Reclamation Systems (01HHE20T)

RELATED ONGOING RESEARCH

Trace Organics Challenge (CEC), Understanding and Communicating Potential Exposure and Effects from Trace Organic Compounds

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Research Outcomes

Through this analysis, the researchers have generated a specific set of gene expression profiles useful as a bioanalytical tool for chemical assessment in reuse waters and other aquatic environments. As with any new tool, the use of microarray analysis will take time to become an established assessment method. These studies were designed as proof of principle for such an application. Given these results and the rapid growth in genomics technologies, the water quality community is poised to evaluate the feasibility of array analysis for assessment of select pharmaceuticals in reuse and other water matrices. This approach will greatly aid in assessment and analysis of potential environmental and human risk in the near future.

Recommendations for Future Research

In the most likely scenario, reuse waters with unknown contaminants will be compared to laboratory waters where background expression values will not interfere with analysis. With further development of arrays and screening of prototypic contaminant classes, training sets can be established to further develop class prediction models for the identification of compounds, or compound classes from complex mixtures of water contaminants. A key to this approach will be the ability to clearly identify non-overlapping gene markers that comprise an established gene expression profiles. This approach will enable a focus on the patterns of gene expression or expression profiles and observe distinct differences in gene regulation. Additionally, if gene annotations are examined, it becomes obvious that specific chemical classes will elicit defined metabolic pathways based upon their mechanism of action.

In an effort to make this technology feasible for water quality assessment, research needs to:

- identify suspect reuse water contaminants by examining urban population pharmaceutical use patterns and demographics;
- develop gene expression training sets by screening of prototypic contaminant classes, in species of interest;
- ascertain dose response effects;
- ascertain time to response effects;
- clearly determine non-overlapping gene markers that comprise an established gene expression profile;
- validate gene expression events by analysis of biochemical pathways based upon known mechanisms of pharmaceutical action;
- determine effects of reuse water matrix on gene expression (TOC, alkalinity, DO, etc.); and
- establish a smaller set of gene expression changes that are indicative of contaminant exposure. Once established, conduct real time polymerase chain reaction (PCR) in place of microarray studies. This analysis will be better suited to the technical capabilities of interested utilities.

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WateReuse Foundation (WRF)

California Urban Water Agencies (CUWA)

The research on which this report is based was funded in part by the U.S. Environmental Protection Agency (U.S. EPA) through Cooperative Agreement No. CR-827345-01 with the Water Environment Research Foundation (WERF). Unless an U.S. EPA logo appears on the cover, this report is a publication of WERF, not U.S. EPA. Funds awarded under the agreement cited above were not used for editorial services, reproduction, printing, or distribution.

01/09