

**REMOVAL OF ENDOCRINE DISRUPTING ACTIVITY FROM WATER RECLAMATION PROCESSES:
COMPARING BIOASSAY RESULTS WITH CHEMICAL ANALYSES**

Jocelyn Hemming*, Jörg Drewes, Sarah Ladenburger Dominick,
Mark Mieritz, Ryan Pieters, James Schauer, William Sonzogni

ABSTRACT: To understand the removal of endocrine disrupting chemicals throughout wastewater treatment, it is important to determine the potential of compounds in wastewater to affect biological activity as well as determine which specific compounds are responsible for the observed biological effect. Therefore, both bioassays and chemical analyses are important tools when assessing the ability of specific treatment processes to remove endocrine disrupting chemicals. We have collected samples from twelve water reclamation facilities throughout the United States and used solid phase extraction to concentrate the samples. Two chemical methods were used to quantify specific chemicals. Enzyme-linked immunosorbent assays (ELISAs) after HPLC clean-up were used to determine testosterone, estradiol and estriol concentrations. GC-NCI-MS was used to quantify several steroidal hormones (testosterone, estradiol, ethinylestradiol, estrone, estriol) and phenolic compounds (bisphenol A, 4-tert-octylphenol, nonylphenol). Estrogenic activity was evaluated using the MCF-7 breast cancer cell proliferation assay (E-screen) and an estrogen receptor (ER) transfected yeast assay. An androgen receptor (AR) transfected MCF-7 cell proliferation assay (A-screen) and an AR transfected yeast assay were used to assess androgen activity. The results of these assays agree reasonably well in assessing the general removal efficiencies of the treatment plant operations. E-screen results from primary treated effluents indicate estrogenic activity ranging from 20 ng/L to near 100 ng/L estradiol equivalents (Eeq). According to the chemical analyses, the vast majority of the E-screen activity is due to three hormones (estradiol, estrone and estriol) rather than phenolic compounds. Results from the ER yeast assay were slightly lower for influent and primary treated samples with the activity due to estradiol and alkylphenols. Testosterone alone was not able to explain all the androgen activity in primary effluents. The ELISA and GC-NCI-MS results agreed well in terms of the substantial removal after secondary treatment, although the specific concentrations were not always consistent.

* Associate Scientist, Wisconsin State Lab of Hygiene, 2601 Agriculture Drive, Madison, WI 53718, USA, Phone: 608-224-6230, Fax: 608-224-6267, Email: hemminjc@mail.slh.wisc.edu