# Report as of FY2006 for 2006ND100B: "Farm-Scale Reconnaissance of Estrogens in Subsurface Waters"

## Publications

- Conference Proceedings:
  - Casey, F., Mary Schuh, G. L. Larsen, Heldur Hakk and Zhaosheng Fan, 2007, "Fate of Manure-borne, Land-Applied Hormones", Third International Water Conference, International Water Institute, Grand Forks, March 13-15.

## Report Follows

# FARM-SCALE RECONNAISSANCE OF ESTROGENS IN SUBSURFACE WATERS

## DESCRIPTION OF STATE OR REGIONAL WATER PROBLEM BEING INVESTIGATED

Endocrine disrupting chemicals (EDCs) are present in animal manures applied to the soil. There is increasing concern on both state and regional levels that EDCs may be leaching to ground water, and that it may be posing a threat to human health through drinking water. Because of this, the fate and transport of EDCs is an important problem of both state and regional concern.

## KEY LITERATURE AND PRIOR WORK

Kavlock et al. (1996) define an EDC as, "an exogenous agent that interferes with the production, release, transport, metabolism, binding, action, or elimination of natural hormones responsible for the maintenance of homeostasis and the regulation of developmental processes in the body." According to Kolpin et al. (2002), natural and synthetic hormones are EDCs that are of particular concern because of their potency and widespread detection. An apparent increase in physiological and reproductive disorders in animals such as fish, turtles, shellfish, gastropods, and mammals has raised concern over the persistence of EDCs in the environment. The effects of these chemicals on human beings remain unknown, but there is concern that evidence of the chemicals' toxicity with animals could be an early indication of dangers that EDCs could present to humans. Disquieting trends, such as increased breast cancer in women and decreased fertility in men have already been linked, in some studies, to EDCs (e.g., Sharpe and Skakkebaek, 1993; David and Bradlow, 1995).

The agricultural use of animal waste as fertilizer is one means of environmental exposure to hormones that has raised concern, because of the intensification of livestock production and crop production near areas of widespread urbanization. Soto et al. (2004) have shown that runoff from concentrated feedlot operations can enter surface waters and result in hormone concentrations that could adversely affect aquatic health. A study by Panter et al. (2000) has attributed the phenomena of vitellogenin production in male fish (*Pimephales promelas*) to increased estrogen levels. Their study showed that vitellogenin production was induced in male fathead minnows that were exposed to 17ß-estradiol for 21 d at concentrations as low as 30 ng/L.

Dr. Francis Casey is my advisor and the project leader of research focusing on estradiol and testosterone and their metabolites at the North Dakota State University. He has conducted past experiments related to the fate and transport of EDCs, and since 2000 he has lead a team of interdisciplinary scientists in a study on the fate and transport of reproductive hormones in the soil. Work by Casey et al. (2003, 2004, 2004) has already been completed to characterize and quantify 17ß-estradiol and testosterone and their metabolites in the soil. Laboratory studies have demonstrated the transformation of 17ß-estradiol and testosterone in natural sediments into various metabolites; however, details of the transformation pathways are still undetermined in the natural environment. It has been found that these hormones degrade readily and are strongly bound to the soil; nonetheless, they are consistently detected in the environment at low concentrations.

The following are related research papers:

Casey, F.X.M., G.L. Larsen, H. Hakk, and J. Simunek. 2003. Fate and Transport of 17ß-Estradiol in Soil-Water Systems. Environ. Sci. Technol. 37(11):2400-2409.

Casey, F.X.M., G.L. Larsen, H.Hakk, and J. Simunek. 2004. Fate and Transport of Testosterone in Agriculturally Significant Soils. Environ. Sci. Tecnol. 38(3):790-798.

Casey, F.X.M., J. Lee, and J. Simunek. 2005. Sorption, Mobility, and Transformation of Estrogenic Hormones in Natural Soil. J. Environ. Qual. 34:1372-1379.

David, D.L., and H.L. Bradlow. 1995. Can environmental estrogens cause breast cancer. Sci. Am. 273:166-172.

Kavlock, R.J. 1991. Overview of endocrine disruptor research activity in the United States. Chemosphere. 39:1227-1236.

Kolpin, D.W., E.T. Furlong, M.T. Meyer, E.M. Thurman, S.D. Zaugg, L.B. Barber, and H.T. Buxton. 2002. Pharmaceuticals, hormones, and other organic wastewater contaminants in U.S. streams, 1999-2000: A national reconnaissance. Environ. Sci. Technol. 36:1202-1211.

Panter, G.H., R.S. Thompson, and J.P. Sumpter. 2000. Intermittent exposure of fish to estradiol. Environ. Sci. Technol. 34:2756-2760.

Sharpe, R.M., and N.E. Skakkebaek. 1993. Are oestrogens involved in falling sperm counts and disorders of the mail reproductive tract? Lancet 341:1392-1395.

Sonnenschein, C., and A.M. Soto. 1998. An updated review of environmental estrogen and androgen mimics and antagonists. J. Steroid Biochem. Molec. Biol. 65:143-150.

Soto A.M., J.M. Calabro, N.V. Prechtl, A.Y. Yau, E.F. Orlando, A. Daxenberger, A.S. Kolok, L.J. Guillette, B. le Bizec, I.G. Lange, and C. Sonnenschein. 2004. Androgenic and estrogenic activity in water bodies receiving cattle feedlot effluent in eastern Nebraska, USA. Environ. Health Perspec. 112:346-352.

Thompson, M.L. 2005. Occurrence, Persistence, and Pathways of 17ß-Estradiol in Agricultural Soils: A Field Study. M.S. thesis North Dakota State Univ., Fargo.

## SCOPE AND OBJECTIVES OF THE PROPOSED RESEARCH

Lysimeters located on a swine (*Sus scrofa*) farm in North Dakota have been installed to monitor the amount of 17ß-estradiol leached through soil, and monitoring wells have also been installed to monitor the surficial aquifer. Based on known application rates in manure and laboratory-determined degradation rates, it was anticipated that most of the hormones would be metabolized or bound to the top 10 cm of soil. Instead, 2003 data (unpublished) show substantial

amounts of 17ß-estradiol leached through the soil and detected in the shallow aquifer. 17ß-Estradiol was even found in the control plot, where no manure was applied, which provided evidence for the antecedent existence of this hormone in the soil or that it was leached from somewhere else.

This project proposes to identify the causes of the unexpectedly high 17ß-estradiol detections. First, the farmer injects his swine waste lagoon material into the field soil at this research location. The soils in this area are aquic and have redoximorphic features (i.e. faint mottling) with 16 cm of the soil surface, which suggests large fluctuations in the water table. Moreover, these soils are derived from water deposited sand, which decreases the sorption of 17ß-estradiol (Casey et al., 2003). We propose the following hypotheses for causes: (1) There exists high levels of background 17ß-estradiol from the lagoon material the farmer injected in the field; (2) High water-table conditions cause a cool and anaerobic environment in which 17ß-estradiol persists; and (3) The ground water that contains the 17ß-estradiol is transporting exogenous hormones into the lysimeters in soil horizon.

## METHODS, PROCEDURES, AND FACILITIES

The research site is a swine farm in North Dakota, where previous and ongoing field studies have and are being conducted by NDSU, concerning the fate and transport of hormones in soil. The farm is an excellent location for the study, because hormone data from lysimeters in four test plots are already available, and six wells are already in place for measurement of water levels and ground-water hormone concentrations.

Both water levels and ground water hormone concentrations will be measured. In addition stratified soil samples will be collected from the surface to the water table, and used to quantify the distribution of the 17ß-estradiol with depth. These findings will be combined with historic lysimeter and well data to examine the spatial and temporal variability of the 17ß-estradiol present in the soil and to determine the causes of its high persistence.

## ANTICIPATED RESULTS AND BENEFITS FROM THE PROPOSED STUDY

It is anticipated that anaerobic conditions will be found in the soil, preventing the degradation of 17ß-estradiol and its metabolites. We also expect to find a correlation between the concentration of 17ß-estradiol in ground water and the height of the water table, and the concentrations of the hormones detected by the lysimeters.

It is hoped that this project will provide valuable information on the behavior of 17ß-estradiol and its metabolites, allowing an assessment of the effectiveness of natural restorative processes in the environment. The proposed ground-water research, combined with data obtained by Casey et al. (2005) on the sorption and mobility of 17ß-estradiol in soil will help to define the interaction of soil and ground water with respect to the retention and transport of 17ß-estradiol. Knowledge of these retention and transport processes may be useful in the development of remediation strategies. Furthermore, since 17ß-estradiol is a prototype for other EDCs, results will contribute to a general understanding of EDCs and other possible organic contaminants.

The proposed project will also benefit society and the environment by serving as an educational tool. Dr. Casey has disseminated his group's findings to the public through several presentations and publications. Past experimental work has been integrated into education

through cooperation with local K-12 students and teachers. A strong effort has been made to incorporate and train individuals from the community and to reach out to underrepresented individuals. Members of the NDSU Soil Sciences department have been involved with school and community programs such as "Extending Your Horizons," which encourages young women in the fields of math and science by fostering awareness about career opportunities related to these studies. They have also participated in "Sunday Academy," an NDSU-Tribal School collaborative project designed to encourage American Indian students in North Dakota to pursue careers in science, math, and engineering. I would like to participate in programs such as these. I believe they would help promote an awareness of the environmental issues that I will be researching. They will also help facilitate interest in the sciences and technology amongst K-12 students and others who, otherwise, might not be given the opportunity to learn from such information.

**PROGRESS TO DATE**

The environmental phenomena to be studied have been identified and we have formulated specific research objectives and hypotheses. I have begun a literature review of the characteristics, properties, and environmental interactions of the chemicals to be studied. Previous local research on the hormones to be researched has been reviewed. I have also begun a literature review on field and laboratory methods for accomplishing the described experimental objectives and will next formulate them into a plan for sampling and analysis.