Pharmaceuticals and Illicit Drugs in Wastewater

Pharmaceuticals can enter streams and underground water reserves through two different pathways. They can be excreted through urine and feces, and unused pharmaceuticals can be disposed at residential areas, hospitals, and veterinary clinics. Although these pharmaceuticals are normally present only in small concentrations, continued consumption through exposure to contaminated water can cause enough bioaccumulation to alter the physiology and biochemistry of animals and humans. In addition, exposure to several compounds simultaneously can cause synergistic and/or antagonistic effects. The process of biomagnification can also occur, causing low concentrations of the compounds in primary producers to increase to large concentrations in predatory species.

Similar to pharmaceuticals, illicit drugs can also enter wastewater effluent through excretion through urine and feces. Through more advanced sampling and analysis, forensic investigators are able to use data collected from wastewater effluent to note the presence of illicit drugs in more specific areas. In Oslo, Norway, scientists were able to analyze sewage samples for ethyl sulfate and ethyl glucuronide, which are found in urine after the ingestion of alcohol, and then estimate the average consumption rates for the region (Reid et al. 2011).

Wastewater Treatment at the Sewanee Utility District

The Sewanee Utility District (SUD) in Sewanee, TN, uses a unique natural wastewater treatment process to serve a small college community. Due to the lack of mechanical components, the SUD has low operational costs. Wastewater enters the treatment plant from Bob Stewman Road and enters a concrete splitter box, which sends the water to lagoons A and B. Each of the three lagoons is clay-lined and covers approximately 4.5 acres each with a depth of 5 feet. In the lagoons, the sewage is treated by aerobic and anaerobic bacteria. Aerobic
bacteria consume oxygen during respiration to convert organic nitrogen to inorganic nitrogen. Algae consume the inorganic nitrogen, and, when they die, they sink to the bottom of the lagoons where anaerobic bacteria decompose organics and convert inorganic nitrogen to dinitrogen. The water stays in lagoons A and B for approximately 30 days and then is moved into lagoon C for approximately 15 days. Time in each lagoon is dependent on the rate of water application to the spray fields. The water is then treated with 8-20 ppm of chlorine after it exits the lagoon to kill any harmful bacteria. This water fulfills EPA and state regulations and is comparable to the water that other wastewater treatment centers discharge straight into the streams. Because the SUD does not have a stream in which they can discharge the effluent, the water is sprayed onto 60 acres divided into 19 spray fields. The water percolates through the soil and is taken up by the vegetation or enters on of three streams: Lab Stream, Stream 6, or Stream 12. As water percolates through the soil, bacteria convert nitrogen so that it can be used by plants, and solid particles, bacteria, and viruses are removed from the water through natural chemical and physical properties. Spraying this water on upland hardwood forests has changed the species composition from drought resistant species (oak and hickory) to more drought intolerant species (yellow poplar and maple). Grasses that form a perennial understory also surrounds the streams from the SUD’s planting wheat, rye grass, and switch grass. These forests receive about 100 inches or precipitation each year, taking into account both natural rainfall and the water from the spray fields.

The population served by the SUD doubles from August to May due to the student population of approximately 1500 at the University of the South. In addition to the increase in population, high rainfall from November to March increases the amount of water that has to be treated. SUD treats twice as much sewage than water is sold, which is due to the large inflow and infiltration problem experienced due to older piping. SUD treats water ranging from 100,000 to 1 million gallons per day depending on the time of year with an average of 300,000 gallons per day.
Study Objectives and Hypotheses

The objectives of this study are (1) to determine what pharmacological metabolites are found in municipal wastewater at the Sewanee Utility District in Sewanee, Tennessee, at different stages of the wastewater treatment process (2) to conduct a seasonal study to determine if the presence of the compounds of interest (especially estrogens) changes when students are present at the university, and (3) to design a greenhouse study in which we observe whether yellow poplars and soil are able to phytoremediate pharmaceuticals from wastewater effluent. We hypothesize that (1) concentrations of compounds identified will be lower in treated wastewater pre and post spray field application than in untreated sewage; (2) estrogens and other pharmaceuticals will occur at greater concentrations during the academic year compared to the summer when the population of Sewanee decreases by about 1500 people; and (3) the combination of yellow poplar and soil do remove some pharmaceutical compounds from treated wastewater effluent.

Methods

Study Site

Samples were taken from the Sewanee Utility District and will be taken from Cookeville’s wastewater treatment facility in the future.

Pharmaceutical Screening Using Passive Samplers

Polar Organic Chemical Integrative Samplers (POCIS) were obtained from Environmental Sampling Technologies. The POCIS samplers are 4 inches in diameter and are composed of two metal rings bolted together with a membrane between the rings. The membrane contains a sorbent that collects any pharmaceuticals present as water flows through
the membrane. The samplers only detect the presence of pharmaceuticals; they do not quantitatively determine amounts.

The POCIS were placed in 5 different locations at the SUD: untreated sewage (sewage chamber), treated effluent before it is sprayed on the spray fields (chlorination chamber), and three streams draining the spray fields. Each POCIS was placed into a stainless steel vegetable steamer, which was wired closed with copper wire. Three POCIS were placed at each location for approximately 10-14 days with a lag time of about 10 days between deployment of the sewer samplers and the deployment of those in the remaining locations. This accounts for the amount of time it takes material to pass through the SUD treatment system. The three POCIS at a single location were combined to a single sample for analysis. At each stream, the POCIS were secured to the weirs, and rocks were placed in front of each weir to maintain water level. The other POCIS were lowered into the sewage and chlorination chambers and secured to the railing.

Passive Sampler Storage

Upon removal from each site, the POCIS were placed in plastic bags and stored on ice. They were immediately extracted at Sewanee.

POCIS Extraction

Cleaning

An aluminum tray was rinsed with methanol and filled with water. Each POCIS disk was placed in the tray and cleaned with a soft brush to remove debris. Care was taken to make sure that the membrane surface was not punctured or contaminated. This step was to ensure that no debris can get into the column while rinsing the sequestration medium.
Extraction

Our work area for the extraction was covered with methanol rinsed aluminum foil. A stopcock was fitted to a syringe with a plug of silane treated glass wool packed into the bottom 2 to 3 cm of the syringe. The syringe with glass wool was used as a chromatography column. The column was then placed onto a holder. A funnel was placed on top of the syringe. We then rinsed to column with methanol and discarded waste. The nuts and bolts were removed from the POCIS disk, and the membranes were separated so that the sequestration medium could be washed into the column using methanol. The rinse portion was collected in a 125 mL beaker. Methanol was allowed to drip through the column so that the rinse portion was raised to a total of 40 mL.

Filtration

A 5mm square piece of filter paper was set into each of the Pasteur pipettes. The pipettes were placed onto a holder with a round bottom flask under each one to collect the sample. The filter paper was wet using a few drops of methanol. The sample collected in the beaker was transferred into the filter pipette and pushed through the filter paper. After filtering, the pipette was rinsed with about 0.5 mL methanol.

Evaporation and Centrifugation

The samples were then evaporated using a rotovap to about 2 mL. To remove any other particulate matter, the samples were ultra-centrifuged at 8000 rpm for 15 minutes at 0° C. Supernatant was transferred into deactivated amber LC-MS vials that were stored at -20° C until they were ready to be analyzed.
Analysis

The samples are then sent to Tennessee Tech University for analysis on a HPLC-QTOF.

Greenhouse Study Design

At the Sewanee Utility District, treated wastewater effluent is sprayed onto a field of yellow poplar trees and then flows into one of the streams that removes water from the area. The purpose of designing a greenhouse study is to observe whether or not the yellow poplar could remove pharmaceutical compounds from the treated wastewater effluent.

To test the phytoremediation of pharmaceuticals, we plan to measure the compounds found in the leachate from pots given water with a known concentration of certain pharmaceutical compounds. There would be three replicated each for pots with yellow poplar saplings and soil, normal upland soil, SUD soil, and effluent only. Each replicate would contain six pots. The saplings in the first set of pots would either be started as seeds, saplings, or cuttings. The next two sets of pots are set up to be a comparison between upland soil and soil that has already been saturated with wastewater effluent. This is to see if soil that has already been exposed to the effluent for an extended period of time retains fewer pharmaceuticals compounds than that which has not been exposed to wastewater effluent. The set with effluent only would be used as a control.

To collect the leachate from each pot, we would construct a system in which the plant would be in a long pot that drained into a bottle that would hold the leachate (Hinchman et al. 1996). We would order pots that were 15 cm wide, 41 cm tall, and would hold a volume of 6.23 L. The bottom these pots would contain a layer of gravel and sand to keep soil particles from getting into the leachate collection bottles. We would need to see how quickly water would drain into the bottles and if exposure to heat in the greenhouse would degrade some of the compounds to determine how often we would need to change the leachate bottles.
Summer 2011 Results

We are currently waiting to see the results of the analysis of our samples that were sent to Tennessee Tech University.

Future Plans

We are now conducting a seasonal study on the presence of certain illicit drugs, pharmaceuticals and personal care products, and estrogens in the wastewater treatment facilities of Sewanee and Cookeville in collaboration with Tennessee Tech University. TTU will analyze the samples for illicit drugs and some pharmaceuticals and personal care products, while Sewanee will analyze the samples for estrogens. In addition to the samples taken in the summer of 2011, we will collect samples in the fall of 2011 and the spring of 2012.

Literature Cited

Hinchman, R et al. “Phytoremediation: using green plants to clean up contaminated soil, groundwater, and wastewater” logos, Argonne National Laboratory (1998)