Analysis of Contaminants in the 13,000 Acres Owned by The University of the South

Walt Doty
Environmental Studies Internship
Professor Shibata
Summer of 2011

Analysis of Contaminants in the 13,000 acres owned by The University of the South

Project Description

The University of the South's campus has become well known not only for the extensive 13,000 acres, but the additional richly diverse ecosystems and wildlife that comprise it. While the human population of the area is relatively small, some parts of the environment have come into direct contact with pesticides and herbicides due to administrative actions of the school, local businesses, and residents. Therefore, the purpose of this study was to investigate various regions of the domain in order to find which contaminants are still present, the identification of any degraded chemicals, and a reasonable estimate of the quantification of theses discovered pollutants.

Thanks to the chemistry department, the utilization of a Liquid Chromatography (LC) as well as High Performance Liquid Chromatography coupled with a Mass Spectroscopy (HPLC/MS and HPLC/MS/MS) was possible. Ideally, the soil samples were separated through chromatography that allowed the identification of specific constituents within each extracted soil solution, and then passed through the MS for the measurement of the molecular weight of the various compounds and their resulting potential degradations. These analytical results were compared with the known standards, thus allowing an understanding of which chemicals are present and to what extent.

Initially our goal was to develop a strategy that permitted the testing of organophosphorus, chlorinated acid, and carbamate pesticides within the soils; however, we realized after sufficient research that the carbamate pesticides would be extremely difficult to extract due to the lacking of certain chemical equipment. We gathered soil samples from various locations which were typically near a water system on central campus. Because of probability that water would of transferred chemicals from nearby regions and relocated in higher concentrations near a stream. Once the soil was collected we immediately extracted the

organopesticides and chlorinated acid herbicides using the methods listed under information for future interns. Unfortunately, the standards that were ordered early on in the research for both organopesticides and chlorinated acid herbicides contained all the chemicals being tested in one vial. Thus, even though we could use the LC to distinguish the chemicals it would be impossible to associate the differing pesticides or herbicides with their corresponding peaks. Even with the addition of the MS, the number of chemicals in the vial were too numerous for isolating the chemical's molecular weight. Thus, with the limited time left in the summer we decided to order only seven independent vialed herbicides and pesticides: dieldrin, picloram, mirex, heptachlor epoxide (isomer B), lindane, 2,4-dichlorophenoxyacetic acid (2,4-D) and dichlorodiphenyltrichloroethane (DDT). We chose theses seven based on the history of Tennessee's and especially Sewanee's environmental intervention as well as the likelihood that these chemicals retained in the region.

The retention times for all of the standards were measured through the HPLC and recorded for reference when be run through the HPLC/MS. Therefore, we could identify with a larger degree of accuracy any contaminants in the soil. However, by the time the standards for all the herbicides finally came in, the summer research period was nearly over. Thus, only three of the seven chemicals were run through the HPLC/MS. The three were picloram, 2, 4 D, and DDT. While we were disappointed with the limited amount of research that capable of being completed this summer, it promising to see the project accelerate much faster near the end. There is still the continuation of testing the remaining vials as well as the already extracted soil samples, and indefinite amount of areas to test for theses pollutants. Thus, this project could be very promising for hopeful researchers.

Information for Future Interns

The procedure for extraction of the Herbicides is listed below:

- Measure out 20.0 ± 0.1 g of soil

- Mix the soil sample with 50 mL of 0.5N KOH in 10% KCl extracting solution (KCl is a solvent). Mix thoroughly by shaking.
- For 15 minutes place the sample into a boiling water bath.
- The sample is transferred to a centrifuge test-tube.
- Centrifuge the sample at 1300 rpm for 15 minutes.
- 3.0mL aliquot is transferred to a small centrifuge test-tube and 150 µl of 12N sulfuric acid is added.
- Vortex the sample for 30 seconds.
- To confirm the pH is < 1.5, pH paper is used. If pH is not below 1.5, additional acid is needed.
- The soil is finally extracted, but needs to be cleaned so 2.0 mL of chloroform is added to the extract.
- This extract should be vortexed and centrifuged at 3000 rpm for 2 min.
- The chloroform layer(lower level) is removed using a pipette and again vortexed and centrifuged for 2 min at 3000 rpm.
- By now the chloroform should have evaporated from the extract and be dry
- Add 4 mL of 1:2 acetonitrille and 0.04 % glacial acetic acid and vortex and sonicate alternately for 5 mins.
- Filter with at least a 0.45 µL the remaining solution in order to prevent the ruining of a HPLC column.

HPLC and MS Starting Recommendations

HPLC*

Flow rate: 0.3 ml/min
Injection Volume: 5-20µl
Mobile phase B: Acetonitrile
Column temperature: not controlled
Column: 2.1 X 150mm
UV range: 230nm
Mobile phase A: 0.04% Glacial Acetic Acid in water

MS*

Injection Volume: 1 µl
ESI (Negative) Drying Gas: 200 ^{O}C
Mass range: 100-600 amu
Gas Flow: 9 L/min
Nebulizer: 40psi
Capillary: 3500V
Skimmer: 35 V

*The information above is by no means the only settings that will gather helpful information. This was just what we found to be the most successful on a routine basis using our equipment.

Personal Experiences

From a personal perspective I found many aspects of this project to be beneficial in the future, but primarily I would like to narrow it down to two specifics. First, I found myself many a time waking up in the morning and not knowing exactly what I had planned for day and there were by no means a certain plan made by somebody else. Thus, I experience a situation that is

very rarely seen in schools or the rest of my life. Second, our project at times was slow moving filled with disappoints. However, it was typically outside of our control, such as the equipment requiring maintenance for a week or perhaps the wrong composition of the standards came in. Therefore, while frustrating it is critical to the success of a long term project to redirect and work on other aspects of the research.

Impact on Career Goals

I discovered that typically I enjoyed the environmental aspects of the project, such as the investigation of the domain, researching the herbicides and pesticides, and other environmental interests. However, the other major component of this project was the laboratory work such as the extractions, working on the HPLC and HPLC/MS, and research within the lab was not nearly as intriguing for me on a day to day basis. Therefore, I think it would be imperative for me to try and keep to jobs that keep me out of the lab and more directly invested with studies such as the environment.

I would also like to thank Sewanee for allowing me to do this research for the summer and Professor Shibata and Asmita Shestha for their work as well.