

STUDENTS BENEFIT FROM INTERNSHIPS

To enhance the international profile of the Network and provide additional training opportunities for our highly qualified personnel, the MITHE-SN was awarded a two-year grant from NSERC's Strategic Network Enhancement Initiative (SNEI). One aspect of this award provided Network students (MSc, PhD, PDF) the opportunity to apply for a short-term internship for 3 months outside of Canada (student exchange or secondment) in a research laboratory, a regulatory agency, an environmental consulting company, or an industry that supplies goods and services related to metals extraction or use. Student internships were approved for 2008 with the Centre for Ecology and Hydrology (CEH) in Lancaster, England; EuroMetaux, Brussels, Belgium; the Centre for Environmental Stress and Adaptation Research (CESAR) and Department of Genetics, University of Melbourne, Australia; and the Nickel Institute, Brussels, Belgium.

Students kept a weekly journal describing their involvement in various activities arranged by their host organization. Highlights of their reports follow.



Kristin Mueller
PhD Candidate

This internship provided opportunities to explore possibilities of modifying the WHAM (Windermere Humic Aqueous Model chemical equilibrium model to take into account spatial or temporal variations in the quality of dissolved organic matter (DOM). WHAM is widely used within the Biotic Ligand Model (BLM) construct to predict the bioavailability, and therefore toxicity, of metals to aquatic organisms in natural systems. Kristin worked on improving our ability to predict the speciation of metals in freshwater, leading to a better understanding of their bioavail-

ability and toxicity. Skills gained were essential for achieving the objectives of Kristin's doctoral thesis.



Julie Sommerfreund
MAsc

The purpose of this internship was to gain an in-depth knowledge of the metal's industry implementation of REACH, the European Union's chemical management system for the Registration, Evaluation and Authorization of Chemicals. During her time at the Nickel Institute, Julie contributed to the technical implementation of several REACH requirements and also had the opportunity to contribute to co-operative programs at EuroMetaux. Through these experiences, Julie gained insight into the application of scientific research to a legislative framework, the use of science based decision making by industry and government, and the co-operation of European industry associations to ensure a consistent approach industry wide to the implementation of REACH. The insights, knowledge, and skills Julie developed will contribute to her efforts at Pollution Probe and the Mining Association of Canada to further develop chemicals management in Canada.

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AQUATIC ECOSYSTEM THEME

Toxicity of selenium in fish – a trout is a trout is a trout ... or is it?



Lana Miller, Ph.D. Student and Will Warnock, Field Assistant, sampling fish from end pit lakes in Alberta in late Fall.

Selenium (Se), an essential element, can become toxic at concentrations only slightly above the homeostatic requirements. Se can cause malformations in fish larvae but less is known about other effects and why some fish are more sensitive than others.

Se can enter the aquatic systems through irrigation of selenium-rich soils, coal mining, and coal burning. As coal mining activities increase to respond to the energy crisis and irrigation expands to provide water for agriculture in many drought-stressed regions, understanding Se toxicology becomes very important.

This project was undertaken to provide new knowledge about sources of Se to the aquatic environments, effects on the physiological status of fish, and the mechanisms underlying different sensitivities of different fish species.

In the first series of experiments, rainbow trout were exposed to Se in the laboratory, and native fish were also sampled in streams located in a mining area and in irrigation canals. Muscle Se was validated as a reliable marker of exposure and effects of Se on the oxidative status of cells were characterized. Next, coal mining end pit lakes with different water Se were stocked with hatchery-reared rainbow trout and brook trout, to be sampled every 6 months. Already at 12 months post-stocking, differences in liver reduced glutathione reserves, thyroid, and adrenal status, growth, and condition between the two trout species were observed. Thus a trout is not a trout...different trout species react differently to Se. Laboratory experiments using head kidney cells from the two trout species are in progress, to determine the mechanisms that make some fish more sensitive to Se, and that make different species of Se more toxic. The study is unique as it will provide long term data from fish exposed to Se in well characterized systems for use in management of Se in the environment. ♣

This research is supported by the Metals in the Human Environment Strategic Network (MITHE-SN).

The research team consists of

Lana Miller (PhD candidate at University of Lethbridge)

Xiaoxi Hu (MSc, University of Manitoba)

Dr. Vince Palace (DFO-Winnipeg)

Dr. Feiyue Wang (University of Manitoba)

Dr. George Sterling (Alberta Environment)

Dr. Alice Hontela and Dr. Joe Rasmussen
(University of Lethbridge)

SOILS & PLANTS THEME

Trace Element Content of Vegetables Grown in Montreal Urban Gardens

Urban gardening is practiced around the world. In some cases, this is the only source of fresh fruit and vegetables for urban dwellers whereas in other cases, it is simply a pleasant pastime. Past human activities have resulted in urban soils becoming contaminated with a wide range of trace elements that may increase the exposure risk of people who eat the food produced.

The objective of his research was to determine if vegetables grown in Montreal urban gardens contain unacceptable concentrations of trace elements. To that end, radish, lettuce, Swiss chard, broccoli, green beans, and tomatoes were grown in 22 urban garden plots. Six additional samples of each vegetable were bought at local food markets and grocery stores for comparison. Trace element content of vegetable tissues and soils, and other soil properties, were measured.

As predicted by the literature, leafy vegetables generally accumulated more trace elements than radish, green beans, and tomatoes. However, there was not much difference between the vegetables grown in the urban gardens compared to those sold in local markets. In some cases, vegetables from the market had higher concentrations than those from urban gardens; for example, tomatoes from the market had significantly higher levels of Cd, Cu, Ni, Pb and Zn, and Swiss chard from the market had higher Cu and Zn. Cadmium in broccoli and Pb in beans were higher in the market vegetables. On the other hand, urban grown lettuce and Swiss chard tended to accumulate more Cd and Pb than samples from the market.

The highest level of Pb was found in Swiss chard with $145 \mu\text{g kg}^{-1}$ fresh weight; beans and tomatoes with $50 \mu\text{g kg}^{-1}$ fresh weight were also found. These levels correspond to half the limit given by the European Union (EU) for these vegetable types and are well below the Canadian Food and Drug regulations that limit the



A productive urban garden in downtown Montreal.

amount of Pb in whole tomatoes to a maximum of $500 \mu\text{g kg}^{-1}$. The level of Cd reached $58 \mu\text{g kg}^{-1}$ fresh weight in lettuce samples, which is far below the EU limit of $200 \mu\text{g kg}^{-1}$ fresh weight for leafy vegetables.

Conclusions

Consumption of urban vegetables can, in some cases, increase the exposure of consumers to trace elements. However, such consumption does not appear to represent risk to human health, and consumption of vegetables enhances human health. ♣

This research is supported by the Metals in the Human Environment Strategic Network (MITHE-SN).

Etienne Foucher was supported by a NSERC USRA.

The research team consists of

Etienne Foucher (undergraduate student)

Dr. Sheila Macfie (University of Western Ontario)

Ms. Marie-Claude Turmel and Dr. François Courchesne

(Université de Montréal)

Dr. William Hendershot (McGill University)

FOODS & INGESTED PARTICLES THEME

Identifying the Real Risk

For humans, ingestion is the main route of exposure to soil-borne contaminants. An important modifier is the extent to which the contaminant is absorbed from the digestive tract – its called bioavailability. Bioavailability can be experimentally investigated by *in vivo* studies or surrogate *in vitro* assays which can range from simple buffered acid solutions to physiologically-based extraction tests (PBETs), which simulate most aspects of a human gut, including the neutral-pH (non-acidic) intestinal phase of absorption (and which may include a simulated intestinal epithelial barrier). The relationship among several *in vitro* assays of bioaccessibility and *in vivo* assays of bioavailability, supported by measurements of mineralogy, is being studied for several field-contaminated soils separated into two particle size ranges. The PBET included digestive enzymes and was two-phase (gastric followed by intestinal); the intestinal phase was supplied to intestinal epithelial (Caco-2) cells grown in monolayers. Total Ni and Pb were measured in both intestinal PBET phase and the cells.

The percent of metal solubilized by PBET in each of the soils that is identified as “bioaccessible” (in other words, available for an uptake by the intestinal epithelial cells) depends on the particle size range:



Figure 1: The rats are gavaged and housed in the metabolic cages for 72 hours (every 24 hours). Metabolic caging is necessary in order to distinguish the truly absorbed metal (gastro-intestinally absorbed, i.e., bioavailable) from the metal excreted from the body in the feces. Urine and fecal output are collected in a separate collection tubes, preventing urine (arrow 1) from washing over and entering the feces tube (arrow 2).

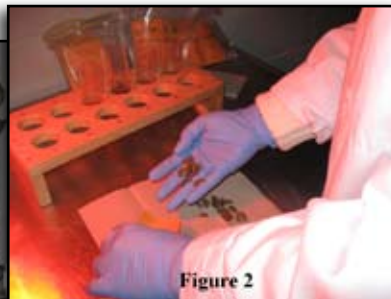


Figure 2: Luba Vasiluk collecting fecal material, while rats remain in the metabolic cages.

Nickel (Ni) and lead (Pb) in the smaller particles are much less soluble in the *in vitro* assays, than the Ni

and Pb from the larger particles. This observation is correlated with total metal in soil, so it's possible that the lower bioaccessibility actually represents a solubility limit to the technique, rather than lower solubility of the metal to digestive processes. Alternately, the speciation (and thus digestive solubility) of Ni and Pb might be particle-size specific, something that the accompanying mineralogy (in progress) will identify. The Caco-2 cells (except for S-1 150-250 μm for Ni) did not absorb all of the Ni or Pb solubilized by PBET as expected; this step represents selective absorption following digestion. *In vivo* studies to validate these assay results are in progress. ♣

This research is supported by the Metals in the Human Environment Strategic Network (MITHE-SN).

The research team consists of

Dr. Luba Vasiluk

Ms. Andrea Amendola

Dr. Beverley Hale (University of Guelph)

Dr. Ken Reimer

Ms. Louise Meunier

Ms. Meghan Lord-Hoyle and

Dr. Chris Ollsen (Royal Military College)

ADVANCING THE SCIENCE OF BIOACCESSIBILITY AND BIOAVAILABILITY IN CANADA



BARC (Bioaccessibility Research Canada) Round-Robin Workshop.

As a follow up to the November 2007 BioAccessability Research Canada (BARC) workshop, a one and a half day workshop was held on September 18-19, 2008. The goal of this workshop was to facilitate information exchange between Health Canada Contaminated Sites Program personnel and the participants of the first BARC Round Robin coordinated by the Royal Military College in Kingston, Ontario. The workshop participants were also drawn from the regulatory, consulting, and academic communities who deal with contaminated sites and are interested in advancing the science of risk assessment through consideration of bioaccessibility and bioavailability.

The progress report from the first round robin exercise was a key discussion item at this workshop. In addition, technical/logistical aspects of a follow up BARC round robin exercise were discussed and planned for the first quarter of 2009. Workshop guest speakers were Dr. Joanna Wragg, British Geological Survey, UK, presenting current Bioaccessibility Research Group Europe (BARGE) initiatives and Professor Nicholas Basta, of the Department of Soil and Environmental Chemistry, School of Natural Resources at the Ohio State University. Professor Basta discussed recent developments in metals bioaccessibility research.

The workshop proceedings will provide important guidance to establish priorities for further research to address data gaps in our understanding of metal bioaccessibility/bioavailability, which will further inform soil bioaccessibility/bioavailability research in Canada and improve confidence for human health risk assessment. Proceedings will be available in early 2009. ¶

Students Benefit From Internships

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Dina Schwertfeger
PhD Candidate

Dina participated in the development of a reference document "REACH Handbook" being prepared to help members of the non-ferrous metals industry find specific information within the REACH guidance documents (RIPs) and also direct members to other metal-specific documents and IT tools available to facilitate the completion of their REACH Chemical Assessment Reports. Dina gained an understanding of the European chemicals policy, REACH, as well as

policies developed at the international (i.e., OECD) level. In particular, she gained an understanding of the organization of the European metals industry, the industry's response to REACH, and the application of scientific research in the policy and the decision-making process. Skills she acquired are applicable to the development and implementation of environmental regulation in North America. The internship has improved her ability to communicate the research from her doctoral thesis as part of the larger frameworks of "environmental quality standards development" and "environmental risk assessment".

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NETWORK NEWS & ANNOUNCEMENTS

Science Communications

The MITHE Strategic Network is now approaching its fifth and final year. The MITHE Board of Directors is keenly aware of the need to link MITHE-supported science to the larger policy and regulatory context in which exposure to metals is assessed and controlled in Canada. To this end, a sub-committee of Board members, with myself as Chair, has been struck to ensure that regulators and policy makers are aware of the insights generated by the research from the MITE and MITHE Strategic Networks.

We are actively exploring mechanisms to make Network research findings more visible and accessible to regulators, policy makers, and to a wider audience which can benefit from the new knowledge.

We will be convening a meeting in January 2009 to stimulate broader discussion about 'policy relevant science' in the context of the MITHE Strategic Network. We will also be developing Discussion Papers which will help prompt further interest in continuing research in this field. We welcome input on methods and approaches to effectively disseminate metals research data to Canadian stakeholders.

Please forward any input or advice to Donna Warner at dwarner@uoguelph.ca.

Alan Penn
Chair, MITHE SN Communications Committee

Board of Directors

We are pleased to announce that Dr. Jim Maguire, Emeritus Research Scientist, Environment Canada, and Mr. Ken Ogilvie, Consultant and Executive Director, Emeritus, Pollution Probe have agreed to remain on the MITHE-SN Board until December 31, 2009.

Biotic Ligand Modelling (BLM) Workshop

MITHE-SN students and researchers participated in the "Biotic Ligand Model Workshop, Principles and Practice" at Wilfrid Laurier University on May 12-14, 2008. The three-day event attracted 60 participants from across North America representing consulting, government, industry associations, and academia. Dr. Jim McGeer, a member of the MITHE-SN Aquatic Ecosystem Theme and his research group organized the workshop, and it was co-sponsored by the Copper Development Association (CDA), the MITHE-SN, the Faculty of Science at Laurier and the McMaster Metals Bioavailability Research Program.

The first day of the program consisted of a BLM short course, led by Dr. Ray Arnold (CDA) and Dr. Bob Santore (HydroQual); it was well attended by students, consultants, and even some experienced modelers wanting to revisit the basics of the BLM approach. Days two and three provided discussions of more advanced BLM principles and a number of MITHE-SN project leaders and co-investigators presented seminars combining research and theoretical considerations (Dr. Uwe Borgmann, Dr. Beverley Hale, Dr. Greg Pyle, Dr. Scott Smith, and Dr. Bernard Vigneault). As well, other leaders in the field of biotic ligand development and application, notably, Dr. Paul Paquin and Dr. Bob Santore (HydroQual), Dr. Paul Welsh (Ontario Ministry of the Environment), Dr. Uwe Schneider and Dr. Patti Gillis (Environment Canada), Dr. Chris Mebane (US Geological Survey), and Dr. Chris Schlekot (Nickel Producers' Environmental Research Association) contributed papers as did a number of students. Vigorous, productive discussions were the order of the day.

NETWORK NEWS & ANNOUNCEMENTS

Patent Award for MITHE-SN Investigator

Dr. Pat Rasmussen was awarded a USA Patent for her Buoyancy-Corrected Gravimetric Analysis System, which she named Archimedes M3™. The new system improves the accuracy of human exposure measurements, and is used for metals and airborne particulate matter samples collected using emergency air monitoring equipment, nano-particle samplers, low-flow PM₁₀ and PM_{2.5} monitors, and personal environmental monitors. The system was custom-designed by Dr. Rasmussen and her team, David MacIntyre and Josée Guénette, to overcome air filter weighing errors caused by changes in relative humidity, air density, particle contamination, electrostatic charge, and physical instability. A customized software/barcoding application automatically calculates buoyancy-corrected mass measurements, providing results in standardized reports. Dr. Rasmussen's facility analysed more than 8000 personal, indoor and outdoor samples from the Windsor Ontario Exposure Assessment Study, and is a key component of her metals research program supported by MITHE-SN and Health Canada.

CNTC Toxicology Short Course

A 10-day Advanced Principles of Toxicology course will be held at the University of Guelph on April 20 – May 1, 2009. This course is organized by the Canadian Network of Toxicology Centres in conjunction with the Centre for Toxicology, the Department of Environmental Biology, and the Office of Open Learning. This university-accredited, open learning course is designed for professionals in industry, government, or education that are currently working or planning to work in areas related to toxicology. Course content will include principles, definitions, and fundamental information designed to provide participants with current knowledge and best practices applied to human health and environmental areas of toxicology. This graduate equivalent course normally requires that learners have a Bachelor's or Master's degree, with course work in statistics, chemistry, and biology. ¶

For registration information on this very popular short course, please contact:

Office of Open Learning: mgaoette@open.uoguelph.ca

On-line Registration: www.open.uoguelph.ca/toxicology

Students Benefit From Internships

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Isabelle Proulx
PhD Candidate

One of the goals of Isabelle's doctoral research project is to determine if it is justifiable to pool *Chironomus* species for contaminant analyses. To achieve this goal, *Chironomus* species needed to be carefully identified. This internship provided Isabelle with the tools and the knowledge to use molecular techniques to identify her species. She also acquired knowledge on identifying *Chironomus* species using both their head-capsules and the giant chromosomes

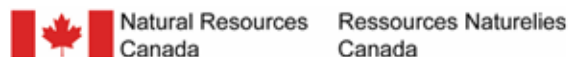
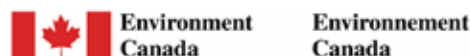
situated in their salivary glands. In addition to being exposed to new ideas, methods and perspectives, Isabelle broadened her network of research connections. Isabelle has stated that she found her internship to be a great opportunity for professional and personal growth. ¶

MITHE STRATEGIC NETWORK SPONSORS

In addition to the sponsors listed below, the Network has research collaborations with 50+ researchers and co-investigators from more than 20 Canadian universities.



Ecotoxicity Technical
Advisory Panel



The MITHE-SN News is a communication produced by the MITHE-SN Secretariat, the Canadian Network of Toxicology Centres, University of Guelph.

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