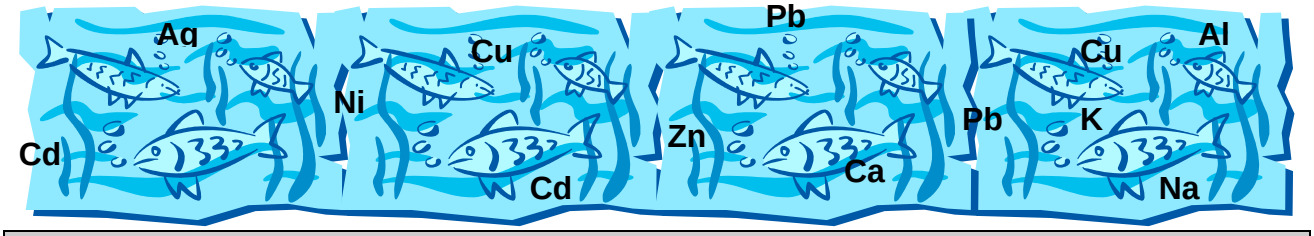
NSERC – Industry Strategic Project on Metal Bioavailability
Research Newsletter



Vol. 7: No. 3 ***McMaster University*** **September 2003**

News

With this issue, Natasha Franklin takes over editorship of our newsletter. We thank Jasim Chowdhury for his excellent work in this position over the past 18 months. Jasim continues in the lab with many other duties, and he is the senior PDF on the NSERC – Industry Strategic Project, replacing Kath Sloman who has taken up a fellowship at Brunel University in London, U.K.

Royal Society of Canada Fellowship for Chris: Chris Wood has recently been inducted into the Academy of Science of the Royal Society of Canada, considered one of Canada's most prestigious accolades. The Royal Society recognized Chris as being a world leader in the area of fish physiology and aquatic toxicology: "He has fundamentally changed our understanding of how fish maintain acid-base balance and regulate internal levels of ions and nitrogen. His research also reveals how such homeostatic mechanisms are perturbed by the effects of acid rain, global warming and metal contaminants. His studies of how metal toxicants act in aquatic environments have changed how regulatory agencies set acceptable environmental levels for toxic metals." Congratulations Chris on this well-deserved honor!

Congratulations also to Eric Pane, who was awarded third place in the 2002 SETAC North America Best Student Platform Presentation Competition for his paper entitled, "The effects of acute and chronic waterborne Ni exposure on growth, reproduction, metabolism, and ionoregulation of *Daphnia magna*."

New PDF in the lab: Fernando Galvez has recently joined our lab as a postdoctoral fellow. This is the second stint at McMaster University for Fernando, who completed his Ph.D. under the supervision of Chris Wood in January 2000. The title of his Ph.D. dissertation was "The Physiology and Toxicology of Waterborne and Dietary Silver Exposure in Freshwater Fish". Since the completion of his Ph.D., he was a NSERC PDF at the University of Alberta with Dr. Greg Goss and a PDF at the University of Waterloo with Drs. Brian Dixon and George Dixon. At McMaster he will be working on mechanisms of metal transport in isolated gill cell populations using techniques he developed while at the University of Alberta.

***BLM accepted by the EU*:** We are pleased to note that the EU and all Member States recently discussed and provisionally approved the use of two critical bioavailability assessment models, the BLM for water and the AVS model for sediments, at the latest Technical Meeting on the Evaluation of Existing Substances (Arona, Italy, June 3-6). Those who contributed, both scientifically and financially, to the development of these concepts and models deserve our sincere thanks. Further information can be found at http://www.eurometaux.org/content.

Research Publications: 2000-2003

Recently we've been asked by funding agencies to document the research output of the Metal's Bioavailability Group over the last few years. We include this list in the current issue as a quick and easy index for readers of the newsletter. Paper reprints of all of the publications listed can be supplied upon request.

2003

- **Chowdhury, M.J., Grosell, M., McDonald, D.G., and Wood, C.M.** (2003) Plasma clearance of cadmium and zinc in non-acclimated and metal-acclimated trout. Aquat. Toxicol. 64: 259-275.
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- **Sloman, K.A., Scott, G.R., Diao, Z., Rouleau, C., and Wood, C.M.,** (2003) Cadmium affects the social behaviour of rainbow trout, *Oncorhynchus mykiss*. Aquat. Toxicol. 65: 171-185
- **Taylor, L., Wood, C.M., and McDonald, D.G.** (2003) An evaluation of sodium loss and gill metal-binding properties to explain species differences in copper tolerance. Environ. Toxicol. Chem. 22: 2159-2166.

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- **Grosell, M., and Wood, C.M.** (2002) Copper uptake across rainbow trout gills: mechanisms of apical entry. J. Exp. Biol. 205: 1179-1188.
- **Kamunde, C., Clayton, C., and Wood, C.M.** (2002) Waterborne *versus* dietary copper uptake in rainbow trout and the effects of previous waterborne copper exposure. Am. J. Physiol. 283: R69-R78.
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- **McGeer, J.C., Szebedinsky, C., McDonald, D.G., and Wood, C.M.** (2002) The role of dissolved organic carbon in moderating the bioavailability and toxicity of Cu to rainbow trout during chronic waterborne exposure. Comp. Biochem. Physiol. C. 133: 147-160.
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- **Taylor, L.N., McGeer, J.C., Wood, C.M., and McDonald, D.G. (2000) The** physiological effects of chronic copper exposure to rainbow trout (*Oncorhynchus mykiss*) in hard and soft water: an evaluation of chronic indicators. Environ. Toxicol. Chem. 19: 2298-2308.

Research Highlights

This issue will highlight research conducted by Eric Pane, a PhD student in the laboratory of Chris Wood. The work was done in collaboration with Dr. Clint Smith and Dr James McGeer, Mining and Mineral Sciences Laboratories, Canada. Results from this work have recently been accepted for publication in *Environmental Science and Technology*.

Mechanisms of acute and chronic waterborne nickel toxicity in the freshwater cladoceran, *Daphnia magna*

Eric F. Pane, Clint Smith, James C. McGeer, and Chris M. Wood

Recent work aimed at the effective regulation of certain metals (Cu, Ag, Cd, Zn, Ni, Pb) in aquatic systems has focused on developing models that will allow for accurate prediction of toxicity of a given metal across a wide range of water chemistries (e.g. the Biotic Ligand Model (BLM) (DiToro et al., 2001). As the focus of such models is the amount of metal accumulated at a specific site within an aquatic organism (e.g. the gill), detailed knowledge of species-specific toxic mechanisms of a metal will enhance the predictive power of such models by providing physiological explanations for the toxicity associated with particular metal burdens. Although recently the acute toxic mechanism (blockade of respiratory gas exchange) of waterborne Ni to rainbow trout (*Oncorhynchus mykiss*) has been elucidated (Pane et al., 2003), neither the acute nor chronic toxic mechanism in *Daphnia* has been previously investigated. The objective of this study, therefore, was to elucidate the acute and/or chronic toxic mechanism of waterborne Ni to *D. magna*.

Acutely, adult *D. magna* were exposed to either control or 694 μg Ni L^{-1} as $NiSO_4$ in dechlorinated Ottawa city tap water (hardness ≅ 45 mg L^{-1} $CaCO_3$) for 48 h without feeding. For chronic exposures, adults were exposed to either control or 131 μg Ni L^{-1} for 14 days (fed

exposure). These concentrations were approximately 65% and 12%, respectively of the measured 48-h static renewal LC_{50} (1068 μg Ni L^{-1}) for daphnid neonates in this water quality.

To test for Ni-related ionoregulatory disturbance, time courses of whole-body ion concentrations were measured, as well as unidirectional influx rates (J_{in}^{ion}) of Na^+, Cl^-, Ca^{2+} and Mg^{2+} using the radioisotopes ^{22}Na, ^{36}Cl and ^{45}Ca and the stable isotope ^{26}Mg.

No impact was observed on the whole-body concentrations or unidirectional uptake rates of Ca^{2+} during either acute or chronic Ni exposure, while only minor effects were seen on Na^+ and Cl^- balance. Chronically, whole-body [Na^+] was significantly reduced (Fig. 1a), though the decrease never exceeded 10%. In comparison to a potent Na^+ antagonist like Ag, these whole-body losses fall far short of the marked whole-body Na^+ losses in *D. magna* reported by Bianchini and Wood (2002, 2003) following acute and chronic Ag exposure.

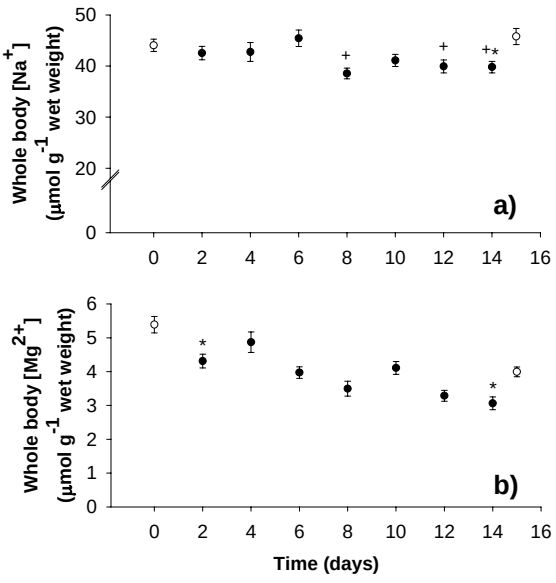

Figure 1: Whole-body ion concentrations in fed, adult *D. magna* exposed for 14 d to 131 μg Ni L^{-1}. Open circles at 0 and 15 d represent initial and final control means, respectively. Dark circles represent experimental means sampled at 2, 4, 6, 8, 10, 12, and 14 d of exposure. n = 8-10 in both groups. (a) [Na^+]. (b) [Mg^{2+}].

The clearest effect of Ni exposure was on Mg^{2+} homeostasis. Acute waterborne Ni exposure antagonized Mg^{2+} homeostasis in *D. magna*, as both whole-body [Mg^{2+}] (Fig. 2a) and $J_{in}^{Mg^{2+}}$ (Fig. 2b) were significantly reduced in exposed animals. Whole-body [Mg^{2+}] dropped linearly with time in Ni-exposed daphnids, becoming significantly reduced by 18% at 48 h (Fig. 2a). $J_{in}^{Mg^{2+}}$ was significantly inhibited by 49% (Fig. 2b) after only 24 h of Ni exposure. Presumably, the problem of Mg^{2+} loss is exacerbated in small crustaceans such as *D. magna*, given very high surface area to volume ratios, lower whole-body Mg^{2+} stores (typically only 25% of whole-body fish Mg^{2+} concentrations), and the lack of a mobilizable Mg^{2+} reservoir in hard tissue such as scale or bone.

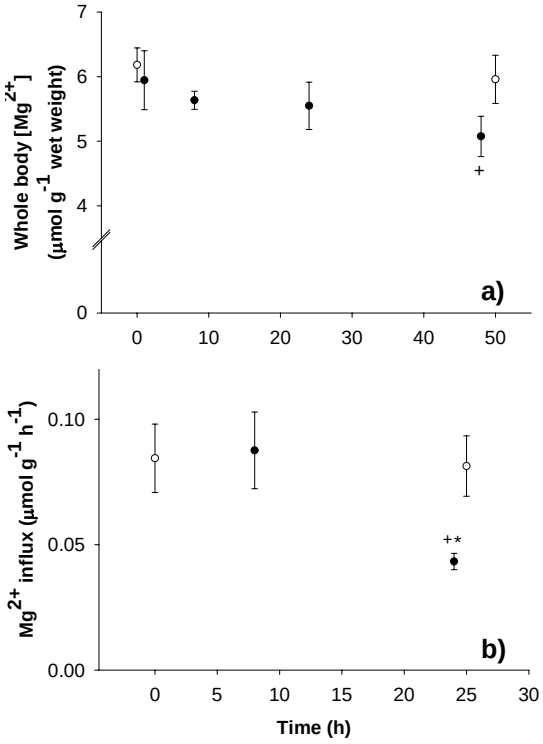

Figure 2: Mg^{2+} homeostasis in starved, adult *D. magna* exposed to 694 μg Ni L^{-1}. Open circles at 0 and 50 h represent initial and final control means, respectively. Dark circles represent experimental means sampled at 1, 8, 24, and 48 h of exposure. n = 8-10 in both groups. (a) Whole-body [Mg^{2+}]. (b) Unidirectional Mg^{2+} influx rates. Note the 24 h time scale on the x-axis.

Additionally, 14 days of exposure to 131 μg Ni L^{-1} caused a significant linear decrease in whole-body [Mg^{2+}] (Fig. 1b), and a significant 47% inhibition of $J_{in}^{Mg^{2+}}$ when measured at the end of the exposure period (0.207 ± 0.016 μmol g^{-1} h^{-1}; control vs. 0.109 ± 0.010; Ni-exposed). At both the first (2 d) and last (14 d) experimental time points, whole-body [Mg^{2+}] was significantly reduced, reaching a concentration at 14 d that was 43% lower than the initial control value (Fig. 1b), although about half of this whole-body [Mg^{2+}] loss was attributable to the holding conditions, as control animals lost 25% of their whole-body [Mg^{2+}] over the same time period.

Although historically, Mg^{2+} has been lumped together with Ca^{2+} under the label "hardness cations," the separate contribution of the two cations to mitigating metal toxicity in aquatic systems has recently been more closely examined (Welsh et al., 2000; Naddy et al., 2002). Welsh and co-authors (2000) found that at a constant total hardness, adjusting Ca:Mg ratios affected Cu toxicity to two salmonid species. Furthermore, Naddy and co-workers found that while higher Ca:Mg ratios protected against Cu toxicity in the rainbow trout, lower Ca:Mg ratios protected *D. magna* (Naddy et al., 2002).

While the specific mechanism of inhibition of Mg^{2+} uptake by Ni is not known, it presents a rich subject for future investigation in *D. magna*. It would be of great interest to examine whether Ni inhibits Mg^{2+} uptake directly via a potential Mg^{2+} channel blockade, or indirectly, perhaps by inhibition of an active Mg^{2+} transporter.

No evidence of acute respiratory toxicity was observed in *D. magna* following acute Ni exposure (Fig. 3a). Neither the rate of oxygen consumption ($\dot{M}O_2$), nor whole-body hemoglobin (Hb) content were significantly affected by Ni exposure (Fig. 3a). Whole body hemoglobin concentrations are known to increase when *Daphnia* experience hypoxic conditions (Peters, 1987). These findings contrast with parallel work from our laboratory with the rainbow trout showing that waterborne Ni elicits acute respiratory toxicity as the mechanism of lethality (Pane et al., 2003). Chronic Ni exposure, however, had a significant impact on both $\dot{M}O_2$ and whole-body [Hb] (Fig. 3b). These two parameters were decreased by 31 and 68%, respectively.

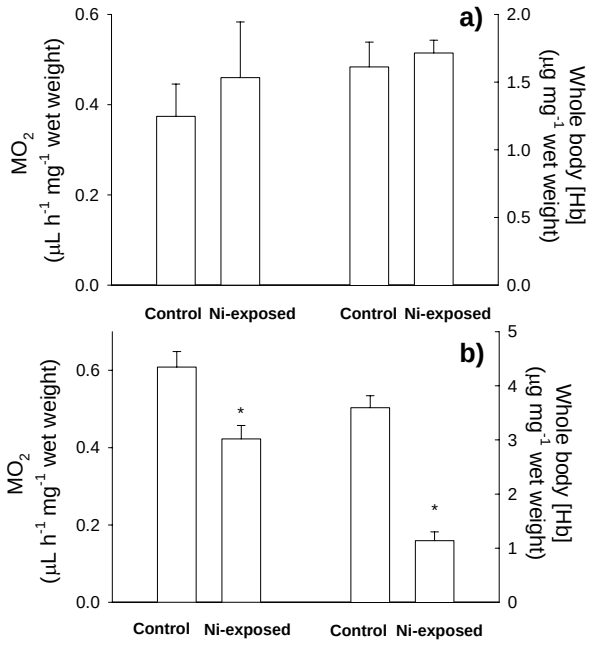

Figure 3: Oxygen consumption rates (MO_2) and whole-body [Hb] content of adult *D. magna*. n = 8-10 in both groups. (a) Starved, adult *D. magna* exposed for 48 h to 694 μg Ni L^{-1}. (b) Fed, adult *D. magna* exposed for 14 d to 131 μg Ni L^{-1}.

Nickel is the second metal, to our knowledge, for which an acute toxic mechanism has been elucidated in *D. magna* – the other being Ag (Bianchini and Wood, 2003). Based on these two studies, it is safe to say that we cannot necessarily assume that the acute toxic mechanism of a metal will be the same for *Daphnia* as for fish. While in the case of Ag, the difference between the two toxic mechanisms is subtle, the difference is marked for Ni (i.e. ionoregulatory in *Daphnia* vs. respiratory in the rainbow trout).

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***Vox Salmonis*:** The lab of Chris Wood hosts a weekly seminar series entitled "*Vox salmonis*." Presentations cover a range of topics in physiology, toxicology, and behaviour of aquatic organisms. We cordially invite anyone who is interested in attending and/or presenting a talk to join "*Vox*" on Tuesdays from 12:00-13:30 on the campus of McMaster University. Please contact Dr. Patricia Gillis (email: gillisp@mcmaster.ca) for more information.

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