IMPACTS PLATFORM ABSTRACTS

Interactive effects of metal mixtures on bioaccumulation and toxicity in *Hyalella azteca*: Are metal mixture impacts "significantly" different than strict additivity?

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Metal mixtures were produced of "equi-toxic" concentrations of each metal. Bioaccumulation of the metals was determined in one-week exposures. Interaction effects were determined by comparison of each metal's bioaccumulation from individual exposures to that in each mixture exposure. A matrix of different mixtures based on seven metals (As, Cd, Co, Cr, Ni, Pb and Tl) were tested in order to determine which metal(s) caused any observed effects on bioaccumulation of any other metal.

Copper, manganese and zinc were not included in the first set of tests since they could confound the results due to partial regulation of copper and zinc by *Hyalella* and also the high concentrations of manganese required to be "equi-toxic". Therefore a second matrix of metal mixtures were tested, based on the seven metal above, in combination with: Cu, Mn and Zn individually, the binary pairs, Cu-Mn, Cu-Zn and Mn-Zn, and finally the tertiary group Cu, Mn and Zn, thus generating the full, 10-metal mixture. Again, bioaccumulation was determined for each metal in order to determine interactive effects on accumulation in one-week exposures.

Mortality rates determined from chronic (4-week) toxicity bioassays were also measured to determine interactive effects on toxicity. A concentration series of "equi-toxic" exposures of the 10-metal mixture was run from which bioaccumulation of each metal and total survival at each treatment was determined. The observed toxicity was less than strict concentration addition based on measured water metal concentrations. Metal concentrations in tissues are nearing completion.

MITE-RN Domain: Impacts, Project C1. Poster Presentation

Assessment of waterborne and dietary factors affecting metal toxicity in fish: towards chronic biotic ligand models (BLMs)

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The biotic ligand modeling (BLM) approach has gained recent widespread interest amongst the scientific and regulatory communities because of its potential for developing ambient water quality criteria (AWQC) which are site-specific, and for performing aquatic risk assessment for metals. The BLM approach is based on an understanding of the key toxic mechanisms of action of metals at the gills, and relates the predicted gill burden in a given water chemistry to the predicted toxic effect. So far BLMs are used for predicting *acute* toxicity (96-h LC50) in any defined water chemistry, however the desirability of using the BLM approach for the assessment of *chronic* toxicity of metals in native species is gaining more or more momentum all over the world, including Canada. The overall long term objectives of our research at McMaster are to use laboratory studies to understand and model the chronic impacts of waterborne and dietary Cu, Cd and Zn on the health of fish (using the rainbow trout as reference) in the environment with the ultimate aim of developing Biotic Ligand Models (BLM) for *chronic* toxicity via both exposure routes. Our present focus is on the effects of different waterborne (e.g. hardness, metal exposure) and dietary (e.g. dietary quantity, quality) factors, which vary greatly in the real environment from place to place and time to time. This presentation will provide an overview of the projects carried out during last year in this aspect, illustrating (i) the effects of dietary Ca and dietary Cd on waterborne Cd and Ca uptake under *acute* Cd exposure; (ii) the effect of sublethal waterborne Cu on voluntary food selection and feeding pattern plus interactions between dietary Na and waterborne Cu uptake; (iii) the interaction between dietary and waterborne Zn uptake; and (iv) the effect of dietary Ca on the responses to waterborne Zn exposure. Moreover, one of the additional aspects of our MITE-RN program is to develop *acute* BLMs for yellow perch (*Perca flavescens*), an endemic fish species in the metal-impacted waters of eastern Canada. This presentation will also include our work on the extension of *acute* BLM approach for Cd to yellow perch in comparison to model species, rainbow trout, and an update on our lab-to-field validation effort related to it. (Supported by NSERC MITE-RN).

MITE-RN Domain: Impacts
Platform Abstract

Direct (physiological) and indirect (food-web mediated) effects of heavy metal exposure on yellow perch

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The biosentinel species, yellow perch (YP: Perca flavescens), was collected from lakes with contrasting metal levels located on the Canadian Precambrian Shield, downwind and downstream from metal smelters in both the Abitibi region and the area around Sudbury. Using a comparative approach, our objectives were to analyse the relationship between metal exposure and (a) ecological factors (habitat quality, food resources), (b) toxicological factors (ambient and tissue metal concentrations) and (c) metal detoxification factors (metallothionein induction and sub-cellular metal partitioning). In lakes at the high end of our exposure gradient, metals (e.g., Cu, Ni and especially Cd) accumulate in YP to concentrations well above background tissue values. Metal accumulation in YP is accompanied by metallothionein induction, but all evidence to date suggests that metal detoxification by metallothionein is incomplete. Indeed, metal toxicity is detected at multiple levels of biological organization, from effects at the cellular level, to effects in organs and tissues, to individuals and populations, in a pattern linked to accumulated metal concentrations (i.e., along the contamination gradient). In addition to direct or physiological effects, we also documented indirect, food-web mediated effects of metals on YP in the most contaminated lakes. The most common indication of such indirect effects on YP is severely stunted growth coupled with a high degree of zooplankton dependence throughout their life.

Population effects of industrial metal contamination in wild yellow perch (Perca flavescens)

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Yellow perch (Perca flavescens) is the only fish species that is ubiquitous in metal contaminated and reference lakes in and around the industrial region of Sudbury, ON. Knowledge of how this species responds to metals in the environment is of considerable importance for ecological risk assessment. Fish were intensively sampled over two seasons, spring and summer, from five lakes of various levels of metal contamination. Crowley, Hannah, and Whitson Lakes are 'contaminated', whereas Geneva and James Lakes are reference lakes. At least 120 fish were captured using various sampling methods during at least two days of sampling in order to sample the widest age range of fish per season. Fish were transported to the laboratory live in aerated native lake water, where fork length, fish weight, liver weight, age, and gender (males, females, and immature) were determined. Fulton's Condition Factor (FCF), Relative Condition Factor (Kn), and hepatosomatic index (HSI) were determined using fish measurement data. Each fish was digitally photographed, and line segments joining superficial landmarks from digital images were used in a truss network analysis to assess ecomorphometric condition. Water and sediment samples were collected at the same time and place as fish capture, and Cd, Cu, Ni, Se, and Zn concentrations were determined using ICP-MS (Testmark, Sudbury, ON). Relationships among muscle anaerobic (using lactate dehydrogenase (LDH)) or aerobic capacities (including citrate synthase (CS), cytochrome c oxidase (CCO), complex I (COMP1)), tissue metal concentrations, environmental (water, sediment, diet) metal concentrations, sampling season, fish size, condition, and age, were also investigated. Fish condition varied by lake, gender, age, and season, highlighting the need for careful interpretation of condition data in wild fish populations. Ecomorphometric components from the truss network analysis were strongly and significantly related to muscle LDH and liver CS activities, and liver protein concentrations; whereas stomach content Zn, liver Ni, liver Cd, and fish condition showed weaker, yet significant, relationships with ecomorphometric condition. Metabolic enzyme analyses suggest that environmental and tissue metal contamination, size and season affect muscle aerobic and anaerobic capacities. However, some indicators of aerobic capacities may be increased in metal-contaminated fish, while others are decreased. From these data, mechanisms of metal toxicity, as well as relationships among diet, aerobic and anaerobic capacities will be discussed. Our study highlights the value of exploiting biological variability along a metal contamination gradient to gain insights into metal-induced effects in wild fish populations, and emphasizes the need to interpret this variability cautiously in light of the confounding influences of season, age, gender, and environmental metal contamination. This research was supported by MITE-RN and by an NSERC Discovery Grant to P. Couture.

IMPACTS POSTER ABSTRACTS

Lake recovery from metal-contamination: the importance of the benthic community for fish energetics

P.G.C. Campbell, Iles, A., and J.B. Rasmussen

The integration of food web ecology into ecotoxicology is proving to be essential. The indirect effects of metal-contamination, through changes in trophic structure, have important consequences for top trophic levels. Living in the 'sinks' of catchments, lake benthic invertebrates are particularly vulnerable to landscape metal-contamination, exhibiting reduced diversity and reduced average body size. Fish in these metal-contaminated lakes are thus subject to the challenge of a simplified food web. Recent research shows the importance of a naturally diverse prey base for maintaining energy transfer to growing yellow perch (Perca Flavescens). As perch grow larger, they shift their diets to larger and larger prey, as it is more energetically efficient for a large fish to eat a few large prey items rather than hundreds of small ones. Otherwise, costs of foraging activity become too high and the fish stop growing. Perch from metal polluted lakes have a very limited choice following zooplanktivory and cannot benefit from the energetic advantages of switching prey. Present research is exploring lakes in different stages of recovery from metal-contamination around Sudbury, ON. Preliminary analysis of benthic samples shows lakes with greater metal pollution have less diverse benthic invertebrate communities and thus more simplified prey bases for benthivorous perch. Perch activity (using lactate dehydrogenase levels as a proxy), condition factor patterns and diet analyses all indicate that heavier reliance on less profitable prey induces energetic bottlenecks and stunting in perch populations.

Metal accumulation in the field: is the diet an important route of exposure for yellow perch (*Perca flavescens)*

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In nature, fish are exposed to essential and non-essential trace metals in both water and food. We measured metal uptake by juvenile yellow perch (YP, *Perca flavescens*) that were caught in a reference lake and held in cages in a lake with elevated levels of aqueous copper (0.06 nM Cu2+), zinc (160 nM Zn2+) and cadmium (3.4 nM Cd2+) in northwestern Quebec, Canada. Twelve floating cages (volume = 5.6 m3) were set up in lakes Dufault (DU, contaminated) and Opasatica (OP, reference). The 0.5 cm mesh size permitted zooplankton, a major source of food for juvenile YP, to freely move in and out of the cages. Juvenile YP caught in OP were either transplanted to DU or caged within their native lake as a control. Between 25 and 30 fish were added to each cage and then sampled at various time points over 30 d. Liver, kidney, gills, digestive tract and carcass were collected for metal and metallothionein analyses. Concentrations of copper and zinc in the organs of transplanted perch were not different from those of control fish after 30 days of exposure, despite the presence of elevated concentrations of both of these metals in the contaminated lake. In contrast, there was a significant increase in cadmium in both the gills and in particular the gut after 30 days of exposure. Surprisingly, Cd levels in the gut of fish transferred to the contaminated lake reached higher levels than those found in indigenous perch in the contaminated lake. The liver and kidneys also accumulated Cd but at a much slow rate than the gut. Our study suggests (i) that under natural conditions YP are able to regulate the uptake of essential metals such as copper and zinc, whereas the uptake of the non-essential metal cadmium is directly related to metal concentrations in the environment, and (ii) that prey are a major source of cadmium for YP under natural conditions. To verify this latter point, we also carried out a pilot feeding experiment in which juvenile YP were collected from Lake OP, caged in small cages (0.24 m3; mesh size 64 m), and fed zooplankton that had been collected either from Lake OP (low Cd content) or from Lake DU (high Cd content). The caged YP were fed 10% of their body weight daily for 20 d, and whole cages were sacrificed at two time points (10, 20 d). Liver, kidney, gills, digestive tract and carcass were collected for metal analysis. Preliminary results suggest that the diet is important for Cd uptake as there were elevated concentrations of Cd in the liver and the kidney following 20 days of exposure to the contaminated diet.In addition to the caging studies, we collected indigenous fish from lakes OP and DU throughout the summer, to track possible seasonal variations in metal concentrations. Results demonstrated no significant seasonal variations in Cd and Zn concentrations in either lake, or Cu in lake DU. Trends were however observed for Cu in the liver and the gastrointestinal tract from fish collected from lake OP: hepatic increased more than two-fold over the summer, ranging from 10  2 microg/g in May to 27  3 microg/g in August; in the gastrointestinal tract, varied more than 10 fold from 1.15  0.15 microg/g in May to 10  1 microg/g in August. These results suggest that seasonal variation of metals should be considered in field biomonitoring programs.

Accumulation and effects of mercury in fish-eating birds

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The general objective of this project is to determine the extent to which environmental mercury is causing adverse effects in wildlife populations in Canada. We determined Hg concentrations in small fish collected from Pinchi Lake, British Columbia, Canada, which in the past received large amounts of waste (primarily HgS) from Hg-mining operations on the shore of the lake; and in fish from several surrounding, uncontaminated lakes. Results indicated that although the average levels of Hg in Pinchi Lake sediments are on the order of 30 times greater than in surrounding lakes, average Hg concentrations in small fish from Pinchi are only a maximum of 2 - 3 times greater, depending on the species, possibly indicating low Hg methylation in this system. Fish from Pinchi Lake bioaccumulate Hg to levels of concern for fish-eating wildlife only once they exceed approximately 7 years of age. We studied Hg exposure and reproduction of bald eagles; while blood Hg concentrations were greater in both adult and young eagles from Pinchi Lake, eagle productivity as measured by nesting success did not appear to be adversely affected. We also did not see a direct relationship between Hg exposure and immunotoxicity, as measured by white blood cell phagocytosis, in common loons having a wide range of blood-Hg concentrations. A good correlation of Hg/Se was, however, observed in various tissues in common loons. Hg and Se speciation was found to be different in different tissues. All Hg in loon eggs were found to be associated with Se. Se may play a role in the storage of de-methylated inorganic Hg in tissues, particularly the liver but the biology of Hg and Se in eggs need further study. Our research is providing new information on the relative risk of elevated methyl Hg exposure for fish-eating wildlife inhabiting aquatic ecosystems.

Ontogenetic changes in dietary and tissue metal concentration in wild yellow perch from a metal contamination gradient

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Previous research by MITE investigators has shown that ontogenetic diet shifts are a normal occurrence in yellow perch. Early in life, yellow perch feed on plankton, but will eventually switch to a diet composed mainly of benthic invertebrates. Large individuals in healthy populations will finally switch to piscivory. Impoverished trophic chains may play a role in the low growth of fish from metal-contaminated environments, in which fish might not be able to switch to larger prey items. Past studies in our laboratory and elsewhere have suggested that diet may be a major source of metal contamination for wild fish. Therefore, the objective of this study was to examine the interrelationships between fish size, diet type (investigated by C3), dietary metal contamination and liver and kidney concentrations of Cd, Cu, Ni, Se and Zn in clean and contaminated lakes. Fish were sampled in spring and summer in five lakes varying in their degree of metal contamination. Crowley, Hannah, and Whitson Lakes are ‘contaminated’ lakes, whereas Geneva and James Lakes are reference lakes. Fork length and total weight were recorded. Fish were then aged and up to four fish from each age class (when available) in each lake were selected for metal analysis. Cd, Cu, Ni, Se, and Zn concentrations were determined in liver, kidney and stomach content using ICP-AES. Fish from the most metal-contaminated lakes (Hannah and Whitson) expressed much higher tissue concentrations of Cd, Cu and Ni than reference fish. Fish pooled from all lakes and ages showed no relationship between Se or Zn concentrations in stomach content and liver or kidney Se and Zn concentrations. However, Hannah and Whitson fish expressed very strong positive relationships between dietary contamination of Cd, Cu and Ni and their corresponding liver and kidney metal concentrations. These data directly support the hypothesis that diet is a major source of metal uptake in contaminated systems. Further analysis examined whether there was evidence of shifts in dietary metal contamination that correspond to shifts in prey types in metal-contaminated lakes. Preliminary results suggest that dietary Cd contamination is very high in young fish, and only decreases in the largest fish sampled in the spring in Whitson Lake. While kidney Cd concentrations reflected dietary contamination, liver Cd remained high even in larger fish. Similarly, Cu concentrations were elevated in the stomach contents and tissues of fish from all sizes, and only decreased in larger fish from Whitson Lake. This study highlights the influence of dietary metal contamination on liver and kidney metal concentrations in polluted lakes. Results indicate that fish from contaminated environments are exposed to elevated dietary metal levels throughout their lives, and that only the few fish that survive to larger sizes may experience a reduction in dietary metal uptake. This research was supported by MITE-RN and by an NSERC Discovery Grant to P. Couture.

Predicting metal and metal mixture effects in aquatic biota.

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Total metal concentrations in the environment do not provide accurate estimates of toxicological effects since toxicity is a function of metal speciation and bioavailability. The main objectives of this project are to determine 1) the best methodology for accurately predicting single metal effects and 2) the most appropriate method of quantifying the effects of metal mixtures. Four studies, all using the benthic amphipod *Hyalella azteca*, and all comparing toxic effects to bioaccumulation, are currently addressing these objectives:

A. Bioccumulation (1-week tests) and chronic toxicity (4-week tests) of mixtures of 7 to 10 metals are being determined. The interactive effects on metal accumulation are being compared with those on toxicity to determine if toxic effects can be predicted more reliably from body concentrations rather than water concentrations. To date, toxicity-bioaccumulation relationships for single metals have been determined for As, Cd, Co, Cr, Cu, Mn, Ni, Pb, Tl, and Zn. Several experiments with mixtures have now been completed; chemical analyses of bioaccumulated metals is nearing completion.

B. A Biotic Ligand Model (BLM) has been produced to explain major ion effects on Ni bioaccumulation and toxicity. The fundamental tenet of the BLM (equal bioaccumulation results in equal toxicity) appears to be valid, at least for 1-week test. Further work will validate the BLM for chronic toxicity, investigate dissolved organic matter effects, and repeat these experiments for Cd. If metal interaction effects are observed in study A, the BLM can be expanded to produce a metal-mixture BLM.

C. The study on the effects of overlying water on metal toxicity in sediments from Sudbury and Rouyn-Noranda area lakes has been completed, and a manuscript is nearing completion. Overlying water has a strong effect on Cd bioavailability, but not on Ni bioavailability because major ion effects on Ni accumulation cancel major ion effects on water-sediment partitioning. The data, based on field-collected water and sediments, will be used to validate the metal-mixture BLM produced under studies A and B.

D. A new study was initiated on bioaccumulation and effects from dietary Cd accumulation. Periphyton are being cultured in the presence of Cd and fed to *Hyalella*. The objective is to determine if Cd accumulated from food is equally toxic as Cd accumulated from water. This has important implications for interpreting the significance of Cd bioaccumulation and performing risk assessments for Cd.

Effects of Cu, Cd and Ni on the physiological status of yellow perch.

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The effects of metals on biochemical, physiological and morphological characteristics of yellow perch, *Perca flavescens*, were investigated in Sudbury, Ontario, in a field study complementary to previous work carried out in Abitibi. Sudbury lakes were selected for this study since some of the lakes are contaminated mainly by Cu and Zn, essential elements, in contrast to Abitibi where Cd, a non essential metal, is the dominant contaminant. To investigate the toxicological effects of Cu, Zn and Cd in perch, six lakes were sampled in Sudbury in June 2002 : two lakes mainly contaminated by Cu and Ni (Hannah et Middle), one lake mainly contaminated by Cd (Crowley), two lakes in recovery (Windy et Richard) et one reference lake (Geneva). The exposure of the fish is characterized by tissue levels of metals (Cd, Cu, Zn, Ni, Pb, As) in kidney and (analyses in progress), as well as metallothionein levels in the liver (analyses in progress). The physiological status of the fish is evaluated with biomarkers of effects, previously tested in Abitibi : secretion of cortisol (plasma levels, *in vitro* production), a steroid hormone important for osmoregulation and intermediary metabolism in teleosts; thyroid hormones, T3 and T4 (plasma levels); activity of gill Na^+/K^+ ATPase, an osmoregulatory enzyme; liver glycogen reserves; histopathological assessment of gills and thyroid tissue; and condition factor as well as hepatosomatic index (indicators of growth).

Preliminary results indicate that perch from lake Hannah, Richard and Middle secrete less cortisol than fish from the reference lake Geneva. Fish from L. Hannah secreted less T3 than fish from L. Richard, Crowley and Middle. Our preliminary results suggest that environmental exposure to Cu and Ni, at the levels found in Sudbury, have deleterious effects on yellow perch. Analyses in progress (biochemical responses, histopathology, metal tissue burdens) will provide data to characterize the physiological status of perch from the Sudbury lakes and the effects of environmental exposures to metals.

The Sudbury field study will be completed by a laboratory study with rainbow trout (*Oncorhynchus mykiss*) that will be exposed *in vivo* to Cu or Zn at a low (environmental) concentration through water and an higher concentration, for 30 days. Same biomarkers of effects as those measured in the field study will be used to furher characterize the effects of Cu and Zn on the physiological status of fish. Since it has been suggested by several studies that exposure to Cu increases the tolerance of fish to other metals, we will also test this hypothesis using our adrenosteroidogenic bioassay *in vitro*. Adrenocortical cells isolated from trout exposed to Cu *in vivo* will be exposed to Cd, Zn or Cu *in vitro* and the secretory capacity of the cells will be determinend. Our field and laboratory studies will provide new data on the physiological effects of metals in teleost fish.

Can metal subcellular partitioning help predict effects in indigenous yellow perch?

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This project is designed to test the prediction that relations between metal accumulation and metal-induced effects will be improved if the metal body concentrations are expressed not as total metals, but rather in terms of the metal's partitioning within a particular organ. We are testing this prediction with indigenous fish (yellow perch: Perca flavescens) collected from 12 lakes with contrasting metal levels (especially Cd, Cu, Zn) but similar trophic status.

Twelve lakes were visited in the Rouyn-Noranda and Sudbury areas in June 2000 and 2001. Water samples were collected by in situ dialysis in the epilimnion of each of these lakes to evaluate ambient metal levels. Juvenile perch (<10 g) were also collected in the 12 lakes and dissected into five body parts: kidneys, liver, gills, gastrointestinal tract and remaining carcass. Each body part was then analysed for metals (Cd, Cu, Zn). Various subcellular fractions of the liver were isolated by differential centrifugation (mitochondria, granules, nuclei and debris, microsomes and lysosomes; heat-sensitive and heat-stable cytosolic proteins) and metal levels were measured in each. Trends in biomarkers (malondialdehyde, glutathione, glutathione-reductase and glutathione-peroxidase,) were then quantified in the liver.

In terms of their relative contributions to total metal body burdens, a shift of metals (Cu and Cd) was observed from the carcass to the liver as metal exposure increased. This result suggests that hepatic Cd and Cu concentrations may exceed the excretory capacity of this organ at higher levels of exposure. The study of metal subcellular partitioning revealed that the liver was not able to cope with all metals in excess. As total metal concentrations increased, the "excess" metal was only partly stored in detoxified fractions in the cells (metallothionein-like proteins of the cytosol, granules); important concentrations were also found in potentially metal-sensitive fractions (mitochondria, microsomes & lysosomes, nuclei). The toxicity of metals associated with these sensitive fractions was tested via a number of biomarkers. The usefulness of metal concentrations in the various subcellular compartments as predictors of cellular toxicity will be discussed.

MITE-RN Domain: Impacts
Poster Abstract

Comparison of the adrenotoxic effects of Cd in Rainbow trout, *Oncorhynchus mykiss*, and Yellow perch, *Perca flavescens*.

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Rainbow trout and yellow perch have a different sensitivity to cadmium (Cd) *in vivo* (trout<LC50<perch). Metals and particularly Cd impair cortisol secretion by adrenocortical cells in both species. The purpose of the present study was to assess *in vitro* the effect of Cd on cortisol secretion by adrenocortical cells of trout and perch, to compare the sensitivity of adrenal steroidogenesis in these two teleosts. Since early life stages of fish are often more vulnerable to contamination than adults, the endocrine toxicity of Cd was also compared in cells of young (1^+) trout and perch.

Cells were exposed to Cd *in vitro* for 60 min, then stimulated with ACTH, dbcAMP, or pregnenolone, a cortisol precursor. Cd inhibited ACTH- and dbcAMP-stimulated cortisol secretion in a concentration-dependent manner in adults of both fish species, however, the EC50s (concentration resulting in 50% inhibition of cortisol secretion) was significantly lower in trout than perch. To test the specificity of Cd to act as an endocrine disrupter, the LC50 (concentration that kills 50% of the cells) was also evaluated to determine the LC50/EC50 ratio.

Adrenocortical cells of adult trout were more sensitive than those of perch and Cd had a higher endocrine-disrupting potential and specificity in trout than in perch. In young fish, the EC50s and the LC50s were not different between adrenocortical cells of young perch and trout. The endocrine-disrupting effects of Cd could be reversed with pregnenolone, maintaining cortisol secretion until cell viability was impaired, in adults and young fish, indicating that site of action of Cd in adrenocortical cells was situated downstream from cAMP formation and prior to pregnenolone synthesis. Our results demonstrate that 1) Cd has a higher endocrine disrupting potential than it is cytotoxic for both species; 2) Cd interferes in the signalling pathway of cortisol synthesis in a step prior to the pregnenolone formation; 3) adrenal steroidogenesis is more vulnerable to Cd in adult trout than perch; and 4) there no differences in vulnerability to Cd of adrenal steroidogenesis between of young trout and young perch.

Effects of sublethal waterborne Cu on feeding pattern in rainbow trout (*Oncorhynchus mykiss*): interactions between dietary Na and waterborne Cu uptake.

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Dietary Na is known to reduce waterborne Cu uptake and accumulation in fish. Therefore, we hypothesize that fish under sublethal Cu stress would prefer Na-enriched food to normal food. We tested this hypothesis in juvenile rainbow trout. Both Cu-exposed (55µg L^{-1} in moderately hard water) and unexposed fish were given the option of selecting between normal and Na-enriched (10% NaCl) food. Four other appropriate control groups were also maintained (normal food & water, Na-enriched food & normal water, normal food & waterborne Cu, and Na-enriched food & waterborne Cu). All fish were fed *ad libitum* and exposed to the respective conditions for 28 days with sub-sampling at days 0, 4, 7, 14, 21, and 28. The diet preference of individual fish was monitored at each sampling time using diets spiked with ballotini glass beads followed by X-ray of whole fish. Apparently Cu-unexposed fish showed preference for Na-enriched food. In the early stages following Cu exposure, both feeding rate and preference were severely affected. Fish recovered from feeding apathy gradually, but the lack of favoritism in food selection was maintained. Growth was severely impacted in all groups of Cu-exposed fish. However, growth and Cu accumulation in target tissues (liver and gill) under waterborne Cu exposure were found to be quite similar in fish given a choice of food relative to those in fish fed with only Na-enriched food. All these responses were significantly different relative to Cu-exposed fish fed with normal diet. Overall, this study indicates that sublethal waterborne Cu exposure affects feeding rate as well as food preference in rainbow trout. Moreover, it indicates that Na-enriched food is beneficial to fish under sublethal waterborne Cu stress but does not fully protect them against adverse effects. (Supported by NSERC MITE-RN).

MITE-RN Domain: Impacts
Poster Abstract