

Impacts of heavy metals on lake food webs: changes to the littoral benthic invertebrate communities and the consequences for yellow perch (*Perca flavescens*).

by

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ABSTRACT

Metal contamination can disrupt the trophic links in food webs by altering the taxonomic composition and size structure of benthic invertebrate communities. Benthic invertebrate samples and perch (*Perca flavescens*) were collected from six lakes along a gradient of metal pollution in Rouyn-Noranda, Canada. The benthic communities of the contaminated lakes were less diverse and had smaller individuals (0.09 mg d.w. and 0.16 mg d.w. vs. 0.22 mg d.w. in the reference lake). The stomach contents of perch from contaminated lakes were less diverse, showed a greater reliance on chironomids or zooplankton, and cannibalism than perch from less polluted lakes. The mean size of prey in perch from contaminated lakes was smaller than in reference lakes (3.7 mg d.w and 7.39 mg d.w vs. 47.7 mg d.w and 67.1 mg d.w. in reference lakes). The mean weight of the largest 25% of the population was significantly higher for reference perch (168.7 g and 89.35 g) than contaminated perch (47.2 g and 24.6 g), which can be attributed to the increased costs of foraging in a simplified prey base. In addition to the effects seen on the growth and diet of perch, we found that the total accumulation of Cd and Cu in the livers of perch was correlated to the proportion of benthivory in the diet of perch.

RÉSUMÉ

Les invertébrés benthiques sont considérés comme proies importantes de plusieurs taxa de poissons. Pour les poissons en croissance, ils représentent une source énergétique transitoire entre l'étape palnctivore et piscivore. La contamination de métaux peut perturber la structure du réseau trophique en changeant la composition taxonomique et la structure de taille des communautés d' invertébrés benthiques. Des échantillons d' invertébrés benthiques littoraux et de perchaudes (*Perca flavescens*) ont été prisent dans six lacs variables dans leur niveau de pollution par métaux lourds dans la region de Rouyn-Noranda, Québec, Canada. Les communautés benthiques provenant des lacs fortement pollués étaient moins divers, et étaient caractérisé par de plus petits individus comparativement aux lacs références (0,09 mg p.s. et 0,22 mg p.s., respectivement). Les contenus stomacaux de perchaudes provenant de lacs contaminés étaient moins divers, caractérisé par une plus grande proportion de chironomides, ou de zooplancton et le cannibalisme était plus commun. La taille moyenne des proies de perchaudes provenant de lacs contaminés était plus petite que ceux des lacs références. Le poids moyen des perchaudes adultes provenant de lacs références était significativement plus élevé que les perchaudes provenant de lacs contaminés. Ceci peut être partiellement attribués aux plus grands couts énergetique associés à l'alimentation dans un système caractérisé par une communauté de proies simplifiées. En plus, nous avons aussi remarqué que l'accumulation totale du Cd et Cu dans les foies des perchaudes est corrélée avec l'importance de l'étape benthivore dans le regime de la perchaude.

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This thesis contains two chapters, each of which has been prepared for publication in a peer-reviewed scientific journal (Canadian Journal of Fisheries and Aquatic Sciences). This thesis represents the results of my own independent research. Both chapters have been co-authored by my thesis supervisor, Dr. Joseph B. Rasmussen. Dr. Rasmussen contributed substantially to the design, execution, analysis, and presentation of the results presented herein. In addition, Dr G. Sherwood contributed to the collection of samples, as well as the theoretical background of both chapters and the original conception of the second chapter. This work was funded by N.S.E.R.C. and M.I.T.E. (Metals in the Environment) grants awarded to Dr. J. Rasmussen.

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GENERAL INTRODUCTION

Energy flows through two primary routes in aquatic ecosystems - pelagic pathways (phytoplankton → zooplankton → fish) and benthic pathways (benthic algae → benthic organisms → fish). Despite this, much of the focus of recent aquatic ecology has been the pelagic pathway (Vadeboncoeur et al. 2002). However, benthic invertebrates are important components of energy flow in aquatic systems. Benthic invertebrate productivity can account for up to 42% of whole-lake secondary production (Vadeboncoeur et al. 2002). Benthic invertebrates also contribute to food webs by decomposing organic matter, oxygenating sediments, and recycling nutrients (Palmer et al. 1997, (Covich et al. 1999) through their various feeding behaviors (ex: filter feeders, biofilm grazers, collectors, shredders and predators). Thus, a diverse benthic community transforms more primary production and allochthonous inputs into useable energy for secondary and tertiary consumers. The diversity of resources used by benthic taxa therefore has the potential to maximize the amount of energy that flows throughout the entire lake food web.

Benthic invertebrates are also important prey items for many fish (Mittelbach & Persson 1998). In a recent review, Vadeboncoeur et al (2002) found that the average North temperate fish species attained 65% of its energy from benthic invertebrates. It is therefore not surprising that fish growth is influenced by the abundance, availability, diversity and size of benthic prey (Mittelbach 1981, Boisclair & Leggett 1989, Heath & Roff 1996, Cobb & Watzin 1998, Hjelm et al. 2000).

Throughout ontogeny, growing fish generally change their niche, which results in changes in their dominant prey type and habitat (Mittelbach & Persson 1998). For example, young-of-the-year yellow perch (*Perca flavescens*) are predominantly pelagic

foragers. By the end of their first summer, perch will move into the littoral zone and start feeding on benthic invertebrates. These type of ontogenetic changes between pelagic and benthic zones are common in freshwater fishes (Mittelbach & Persson 1998). These ontogenetic changes are also associated with increased size of prey. Keast (1977) found that as yellow perch increased in age, the dominant size of prey increased progressively from 0.1 mg to 100-250 mg in the oldest age class of perch. Similarly, yellow perch growth rates were negatively correlated with the percent contribution of small prey in their diets (Boisclair & Rasmussen 1996). According to theoretical models of fish growth, foraging costs increase when predator size increases relative to its prey, resulting in decreased foraging efficiency (Kerr 1971). Benthic invertebrates are therefore, an important dietary stepping-stone between zooplanktivory and piscivory, as they span a wide range of sizes (e.g. < 0.5 mg – 512 mg) (Hanson et al. 1989, Rasmussen 1993). Having diverse prey sizes in a food web allows omnivorous fish to meet their energetic demands as they grow and helps to increase trophic efficiency by lowering the cost/benefit ratio of their foraging (Werner & Gilliam 1984, Sherwood et al. 2002b). For fish that have a benthivorous stage during ontogeny, the absence of large benthic prey can result in stunted fish (Heath & Roff 1996, Sherwood et al. 2002a). Therefore, any perturbation to the food web that results in the loss of all or part of the benthic invertebrate community has the potential to severely alter how efficiently energy is acquired by higher trophic levels.

Many aquatic ecosystems in North America are chronically exposed to heavy metal contamination (Luoma 1989). Consistent patterns of taxonomic change in chronically metal polluted environments are observed in the benthic invertebrate

communities of experimental systems (Winner 1975, Leland 1989) and natural systems (Johnson et al. 1992, Gower et al. 1994, Day et al. 1995, Poulton et al. 1995, Clements et al. 2000). In these systems, the abundance of metal-sensitive benthic taxa (e.g. most ephemeropterans, odonates and gastropods) is reduced and the community becomes dominated by fewer, metal-tolerant taxa (e.g. chironomids). It is important to note that those taxa that are typically considered sensitive to heavy metal contamination are generally taxa that form the majority of the large end of the benthic size spectrum. Thus, high taxonomic diversity in benthic invertebrate communities will help maintain a wide range of prey types, quality and prey sizes.

In chronically polluted systems, the benthic invertebrate communities become dominated by smaller bodied individuals relative to reference systems (Gaston et al. 1998, Carlisle 2000). Thus, one of the important impacts that heavy metal exposure can have on food webs is to shift the available benthic prey resources towards a less diverse prey base, with a smaller mean invertebrate size, which can compromise the efficiency of energy transfer through the benthic pathway. This can manifest itself as an alteration in the diet patterns of benthic feeding fish. In metal-polluted systems, fish can have simplified stomach contents (Munkittrick et al. 1991, Clements & Rees 1997), higher than normal rates of zooplanktivory, decreased growth/consumption rate (conversion efficiencies) (Sherwood et al. 2000) and increased foraging costs (Sherwood et al. 2002a). The various increased costs of foraging in simplified prey bases can potentially account for the reduced growth of higher trophic levels (Sherwood et al. 2002a). Therefore, by preventing fish from following through their normal ontogenetic habitat

and prey shifts, metal contamination can effectively uncouple the linkages between benthic and pelagic pathways at higher trophic levels.

Although switching to benthivory offers important energetic benefits to many fish, it can also potentially increase a fish's exposure to metal contamination. Sediments often contain the highest loading of metals in aquatic ecosystems (Luoma 1983). As a result of living in, and feeding on, contaminated sediments, benthic invertebrates tend to have high concentrations of heavy metals (Smock 1983, Farag et al. 1998). Thus, benthic invertebrates can also form an important link in the trophic transfer of metals in freshwater systems (Dallinger & Kautzky 1985, Munkittrick et al. 1991, Woodward et al. 1994, Farag et al. 1999).

Fish can accumulate metals from their diet at each stage of their life, but the concentrations of metals in their diet may not be the same at each stage. Pelagic food webs have inefficient trophic transfer of metals (Baines et al. 2002). Furthermore, zooplankton elimination rates of metals can also be up to an order of magnitude higher than some benthic invertebrates (Reinfelder et al. 1998). Conversely, benthic invertebrates have mechanisms for storing metals in or on their bodies (Hare 1992). Therefore, as a fish like perch changes its diet from a primarily zooplanktivorous diet to benthivorous diet, it can potentially increase the total amount of metals to which it is exposed. Changes in metal accumulation associated with ontogenetic diet shifts have previously been reported for marine turtles (Sakai et al. 2000) and in chronically exposed freshwater fish, benthic feeding stages have been identified as survivorship bottlenecks (Amundsen et al. 1997, Farag et al. 1998, Farag et al. 1999). However, to our

knowledge, no study has specifically studied diet changes as a potential mechanism for changes in metal exposure.

The focus of most ecotoxicological work has been lab based and population or individual based (Munkittrick 1995). Although this information is a necessary precursor to having a complete understanding of contaminant impacts, without a clear understanding of the higher order ecological impacts, we will not be able to adequately protect, manage and restore contaminated systems. The purpose of this thesis is to explore some of the impacts of metal contamination on aquatic food webs, with a specific focus on the role that changes in benthic invertebrate communities has on higher trophic levels.

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CHAPTER 1: Impacts of altered benthic invertebrate communities on the feeding ecology and growth of perch (*Perca flavescens*) in metal contaminated lakes.

Introduction

Chronic pollution from heavy metals is widespread in the environment. Generally, ecotoxicologists assess the impact of these pollutants by using standard toxicological assays (e.g. L.D. 50's) or by relating metal concentrations in tissues of test species, or some physiological endpoint (e.g. inductions of enzymes, individual growth) to metal concentrations in the surrounding environment. An alternative approach has been to measure changes in the community structure among sites with different degrees of pollution. Benthic invertebrate communities are considered one of the most sensitive indicators of perturbation from heavy metal pollution in aquatic ecosystems (Reynoldson 1992). Consistent patterns of taxonomic change in chronically metal polluted environments are observed in the benthic invertebrate communities of both experimental (Winner et al. 1975, Clements et al. 1989) and natural systems (Leland et al. 1989, Johnson et al. 1992, Gower et al. 1994, Day et al. 1995, Poulton et al. 1995, Clements et al. 2000). In these systems, the abundance of metal-sensitive benthic taxa is low and the communities are dominated by fewer, metal-tolerant taxa.

Reductions in taxonomic diversity in unpolluted systems have been linked to decreases in various ecosystem functions such as community respiration, productivity, and decomposition (Naeem et al. 1995), biomass production (Pfisterer & Schmid 2002), and increased consumption rates (Cardinale et al. 2002). However, researchers studying the effects of heavy metal pollution on aquatic invertebrate communities have limited their analysis to changes in taxonomic composition. Relatively few studies have examined how the reduction of taxonomic diversity in pollutant-exposed benthic

invertebrate communities results in disrupted food web structure (Munkittrick & Dixon 1988, Clements & Rees 1997).

The reduction of taxonomic diversity in chronically metal-exposed benthic communities can affect the functional attributes of food webs and communities (Gaston et al. 1998). Benthic invertebrates are important in aquatic systems for decomposing organic matter, oxygenating sediments, and recycling nutrients (Palmer et al. 1997, Covich et al. 1999). A wide range of invertebrate feeding behaviours (filter feeding, biofilm grazing, collecting, shredding and predation) can transform more of the available primary production and allochthonous inputs into useable energy for secondary and tertiary consumers. This diversity of resources used by benthic taxa can help maximize the amount of energy that flows to the top of the food web. In a recent review of the literature, Vadeboncoeur et al. (2002) found that for 15 species of north temperate lacustrine fish, the percent reliance on benthic invertebrate derived prey ranged from approximately 20% to 100%, with the average species attaining 65% of its energy from benthic invertebrates. Thus benthos can be a dominant source of energy for top predators and also represents an alternative or complementary energy pathway other than pelagic, zooplankton energy.

Growth of top predators, like fish, is probably one of the most common indices of bioenergetic health in studies of pollution (Langston 1995). Although fish toxicologists often relate poor growth to effects of direct toxicity (toxins impair physiological functioning, which in turn prevent fish from maximizing energy allotted for growth) (Munkittrick 1995), fish growth can be indirectly affected through alterations in the type and diversity of benthic invertebrates (Clements & Rees 1997). The abundance,

diversity, and size of benthic prey influence fish growth (Mittelbach 1981, Boisclair & Leggett 1989, Heath & Roff 1996, Cobb & Watzin 1998). Surprisingly, few studies have examined the influence of pollution-induced changes in prey availability on fish foraging habits (Munkittrick et al. 1991, Clements & Rees 1997). The impacts of disturbed benthic invertebrate communities can also manifest themselves as an alteration in the diet of benthic feeding fish. Perch in metal contaminated lakes have increased foraging costs (Sherwood et al. 2002) and decreased conversion efficiencies (Sherwood et al. 2000). These changes might be a direct consequence of perch foraging in less diverse benthic communities with reduced availability of larger prey items. Thus, high taxonomic diversity in benthic invertebrate communities may also ensure that a wide range of prey types, quality and perhaps most importantly, prey size are maintained in a food web.

Body size influences many of the important roles performed by benthic invertebrates. The distribution of body sizes within benthic invertebrate communities affects the fate and rate of benthic production and processes (Strayer 1991). Thus, size distributions of benthic invertebrates might affect the efficiencies of energy transfer and the growth of fish. The benthic invertebrate taxa that are most sensitive to metal pollution tend to be those that attain large maximum biomass. The greater susceptibility of certain taxa tends to be related to the feeding habits of those taxa (Smock 1983). In chronically polluted systems, we would expect benthic invertebrate communities to be dominated by smaller bodied individuals relative to reference systems that should have more large individuals (Gaston et al. 1998, Carlisle 2000). Reduced size distributions in benthic invertebrate communities may have important implications for higher trophic levels that are dependent on benthic resources for energy.

Switching to progressively larger prey items is a common strategy of freshwater fish (Mittelbach & Persson 1998). As such, benthic invertebrates are an important dietary stepping stone between zooplanktivory and piscivory. Having a wide array of prey sizes in a food web allows omnivorous fish to meet their changing energy demands as they grow and helps to increase trophic efficiency by lowering the cost/benefit ratio of foraging (Boisclair & Leggett 1989, Sherwood et al. 2002). Lack of appropriate prey sizes can potentially lead to stunting in fish (Heath & Roff 1996, Sherwood et al. 2002). From the perspective of an omnivorous fish, it is not necessarily the taxonomic identity of prey that is crucial for attaining maximum body size, but the availability of appropriate prey size that allows fish to reach their full growth potential.

In this study, we sought to explore some of the effects of metal exposure on food webs in contaminated systems. We demonstrate that reduced taxonomic diversity of chronically exposed benthic invertebrate communities is associated with a reduction of their size distributions. Furthermore, we demonstrate that disrupted invertebrate community structure is linked to alterations in the diet patterns of a common freshwater fish, the yellow perch (*Perca flavescens*). Lastly, we discuss how these changes have led to impaired growth in yellow perch.

Methods

Study area – The city of Rouyn-Noranda, in north-western Quebec, Canada, has a long history of mining activity that has led to the accumulation of heavy metals in the water column and sediments of lakes downwind from the city's copper refinery (Wang et al. 1999). Lakes upwind from the refinery have dissolved and sediment metal concentrations that are low relative to those in lakes downwind from the refinery (Fig. 1). Six study lakes were chosen to represent a gradient of Cu and Cd contamination and a narrow range in limnological characteristics (Table 1). Two lakes upwind from the refinery, Opasatica (OP_R) and Dasserat (DA_R), are reference sites. Two lakes were chosen as high pollution sites, Osisko (south basin) (OS_P) and Dufault (DT_P). OS_P was the mine-tailing pond for the refinery. However, it no longer receives tailings. DT_P has received mine drainage and atmospheric deposition of metals from the refinery. Finally, two lakes were considered to have intermediate levels of pollution, lake Vaudray (VA_I) and Bousquet (BO_I). Both these lakes are downwind from the refinery and have only received atmospheric deposition of metals.

Benthic invertebrate community sampling and identification – Benthic invertebrates were collected from study lakes in June 1999 using a kicknet and Eckman samplers. We collected samples from four different sites at three depths (1m, 2m and ≥ 3 m)/ site in each lake. At each site, one sample was taken within a macrophyte bed (if present) and one was taken outside of the macrophyte bed for six samples/site. Samples were sealed in plastic bags, kept on ice in the field, taken back to the lab and frozen. We made no special effort to sample crayfish and unionid bivalves in this study, although the presence

of unionid bivalves has been documented for the intermediate and reference lakes (Wang et al. 1999). Benthic invertebrates were identified to order for all lakes and to family for a subset of lakes, using a dissecting microscope. All individuals within sites were counted and then identified using published taxonomic keys for freshwater invertebrates of North America (Peckarsky et al. 1990).

Fish sampling – Yellow perch were collected in June of 2000 in the evenings from each lake. Methods of capture included gill nets and hook and line. Young of the year (Y.O.Y.) were collected using a Mysis net that was towed within and just outside of the littoral zone. We selected approximately 30-100 fish from as wide a range of fish sizes (2 – 30 cm) as possible. Perch were put on ice in the field, and immediately frozen upon return to the lab.

Benthic invertebrate community diversity analysis – Shannon-Weaver's index of diversity was used to compare average benthic community diversity among lakes. Shannon-Weaver's index was calculated as:

$$(1) \quad H' = - \sum p_i \log_2 p_i$$

Where $p_i = n_i/N$ and n_i = number of individuals of the *i*th taxa and N = total number of individuals. We used H' to assess the diversity at the level of order and family.

Invertebrate Size Analysis – Total length and head capsule width of aquatic insects and hydrachnids (bivalves and gastropods were excluded from size analysis) from OP_R, OS_P, and DT_P, were measured using an ocular micrometer and dissecting microscope. For each site and for each taxon, pooled or individual invertebrates were placed in pre-weighed tin capsules and dried for 24 hours at 70 °C. Complete individuals from each taxon were pooled according to total body length (e.g. ≤ .5, .51- 1, 1.1- 1.5 cm, etc.). Dried invertebrates were weighed using a microbalance and average weight/individual was determined for pooled samples. Relationships between head capsule width, total length, and body weight were established to determine estimates of weight/individual for those individuals that had incomplete bodies.

Perch Diet Analysis – We used gut content analysis and stable isotope analysis to establish the diet of perch. Perch were thawed, and the operculum, stomach, and a small piece of dorsal white muscle was removed. Opercula were used to identify the age class of perch. For each fish, we measured the total number of individuals of each taxon and the average size of prey in the stomach contents. Diet diversity was measured as percent occurrence (percentage of fish/age class/lake with a particular prey item in their stomach contents) (Hyslop 1980, Marshall & Elliott 1997). We used stable isotopes to establish a more integrated measure of diet patterns. Stable ¹³C signatures of consumers generally reflect the ¹³C signature of their prey. Therefore, ¹³C signatures are conserved from the bottom of the food web to the top (Deniro & Epstein 1978, Vander Zanden & Rasmussen 1999) and can provide information about the original source of carbon within a food web. Consumers become enriched in ¹⁵N relative to their prey by 3-4‰ (mean 3.4‰) (Deniro

& Epstein 1981). Therefore, stable ^{15}N signatures indicate trophic position within a food web (Vander Zanden et al. 1999b). Zooplankton and benthic invertebrate samples were collected in June 2000 to determine prey ^{13}C and ^{15}N signatures. Zooplankton was collected by conducting horizontal and vertical tows in the epilimnion with zooplankton nets (mesh size: 250 μm). Littoral benthic invertebrates were collected using kicknets and profundal benthic invertebrates were collected using Eckman samplers.

Whole samples of invertebrate, pooled zooplankton and perch muscle tissue were frozen, dried at 70 °C for 48 hours in a drying oven and ground into a fine powder using a mortar and pestle. Isotopic content was measured using a mass spectrometer at the G.G. Hatch Laboratories, University of Ottawa. A more positive signature is isotopically enriched and contains more of the heavier isotope. Trophic position was calculated as:

$$(2) \quad \text{TP} = ([\delta^{15}\text{N}_{\text{consumer}} - \delta^{15}\text{N}_{\text{baseline}}]/3.4) + 2$$

Where $\delta^{15}\text{N}_{\text{baseline}}$ is the ^{15}N signature of the average herbivorous littoral invertebrate (trophic position 2) for each lake. Baseline signatures are used to represent the trophic position of a consumer relative to the $\delta^{15}\text{N}$ of a primary consumer (Vander Zanden et al. 1999b). We used the average littoral invertebrate $\delta^{15}\text{N}$ as the baseline indicator. 3.4 represent the average ‰ increase per trophic level.

Results

Benthic invertebrate diversity - Total benthic invertebrate diversity at the level of order, as defined by the Shannon-Weaver index (H'), was greatest in the reference (OP_R and DA_R) and intermediate lakes (VA_I and BO_I) (Fig. 2a). The polluted lakes had the lowest diversity (One-way ANOVA and Tukey-Kramer multiple comparison test: $F_{5,79}=11.049$, $p = <0.0001$). The differences in H' are attributable to the taxonomic differences among

lakes in the mean abundance/m² of several taxa (Table 2). One-way ANOVA and Tukey-Kramer test comparing the mean abundance/m² within taxa, among lakes revealed significant differences between at least one reference lake, usually DA_R, and either one of the polluted lakes, or the polluted lakes and the intermediate lakes, in amphipods, gastropods, hemipterans, hydrachnids, and trichopteran (Table 2). Although the mean abundances of ephemeropterans and odonates were greater in the reference lakes, the variability in the data was too great to detect a significant difference among lakes.

At the level of family, the patterns of diversity were similar (Fig. 2b) for OP_R, DT_P, and OS_P (the only lakes with family data). OP_R had a significantly higher mean H' than OS_P or DT_P (One-way ANOVA and Tukey-Kramer multiple comparison test: $F_{5,79} = 10.934$, $p=0.0003$). There were no significant differences between the two polluted lakes.

Benthic Invertebrate Biomass – Biomass of benthic invertebrates was measured for three of the study lakes, OP_R, DT_P, and OS_P. Size analysis revealed differences among the three lakes (Table 3). However, the difference in biomass was only significant between OP_R and DT_P (One-way ANOVA, $F_{2,1724} = 8.88$, $p = 0.0001$, followed by Tukey Kramer test). There were no significant differences in biomass between OP_R and OS_P. The larger individuals in OP_R tended to be odonates, hemipterans, trichopteran and ephemeropterans, whereas in the contaminated lakes, dipterans were the only large individuals.

Perch Diet – Stomach Content Analysis – The stomach content data broadly separate the six lakes into two groups: 1) OP_R, DA_R, and BO_I – typical perch ontogenetic diet patterns and 2) VA_I, OS_P, and DT_P –deviant ontogenetic diet patterns. In OP_R and DA_R, the

general diet patterns indicated a brief planktivory stage (Table 4). In OP_R, 35% of 1⁺ perch had zooplankton in their stomach contents. 1⁺ perch also had dipterans in their stomach contents, but at a lower proportion (Table 4). In DA_R, only one 1⁺ perch was caught. Therefore, the 1⁺ and 2⁺ age class was grouped together. Twenty-two percent of the 1⁺ and 2⁺ perch had zooplankton in their stomach contents. There were more 1⁺/2⁺ perch (55%) in DA_R that had dipterans in their stomach contents than zooplankton. This is probably due to the lack of 1⁺ perch and not a reflection of an absence of planktivory. The benthivorous stage of reference perch had high taxonomic richness and generally, the stomach contents of older perch contained benthic taxa that attain larger maximum sizes (e.g.: odonates and crayfish) (Table 4). OP_R perch had less taxonomic richness than DA_R (6 orders of benthic invertebrates in OP_R vs. 10 in DA_R). Benthic invertebrates dominated the stomach contents of 2⁺ - 4⁺ perch in OP_R. This was true for all age classes of perch in DA_R. OP_R perch were the only ones from all study lakes that demonstrated sustained piscivory (increasing in dominance through more than one age class) (Table 4). Fish was the third most important prey item for 4⁺ perch in OP_R, and the most important for subsequent ages. The perch in DA_R did not show any sustained piscivory, but did begin eating crayfish at 2⁺.

The perch from the intermediate lakes (VA_I and BO_I) showed similar general patterns as DA_R, a planktivorous stage followed by a mixed benthic stage and no full switch to piscivory (Table 4). VA_I perch had less benthic taxonomic richness in their stomach contents than BO_I or the DA_R (6 orders vs. 10 for BO_I and DA_R). *Hexagenia*, a large ephemeropteran, was the dominant prey item in all age classes for VA_I perch (Table 4). In BO_I, *Hexagenia* was present in all age classes and was a primary diet item by age

2⁺. Amphipods were also a major prey item for 1⁺ and 2⁺ perch in BO_I (Table 4). By 4⁺, BO_I perch were feeding on crayfish in addition to *Hexagenia* and other large bodied invertebrates like odonates.

The polluted lakes (OS_P and DT_P) showed different dietary patterns, both from the less polluted lakes and from each other (Table 4). OS_P perch did switch to benthivory at 2⁺, but the benthic portion of their diet had fewer prey taxa than perch from less polluted lakes. OS_P perch fed almost entirely on dipterans (mostly Chironomidae larvae and pupae) and to a lesser degree, trichopterans (mostly Leptoceridae). There was no shift to benthic taxa that achieve large maximum size and no sustained switch to piscivory. Unlike the perch from all the other study lakes, a high proportion of DT_P perch (100% - 44%, age 1⁺ - >5⁺) had zooplankton in their stomach contents. In addition, the benthic prey items were dominated by dipterans (40% - 82% occurrence, age 1⁺ - >5⁺). The shift from a diet dominated by zooplankton to one dominated by benthic invertebrates did not happen until 4⁺ in DT_P perch.

The average mass of non-zooplankton prey items (excluding 1⁺ perch) in the stomach contents of perch from reference and intermediate lakes was significantly higher than perch from the polluted lakes (Fig. 3, one-way ANOVA and Tukey-Kramer multiple comparison test, $F_{5, 173} = 5.94$ and $p < .0001$). The prey in reference perch were approximately double the size on average than the prey in perch from intermediate lakes and approximately 7 – 20 times greater in size on average than prey in OS_P or DT_P perch (respectively). In general, as perch from reference and intermediate lakes got older, the mean size of prey items (excluding zooplankton) increased (Fig. 4). This was different

from the polluted lakes where there was virtually no change with age in the mean size of prey (Fig. 4).

Perch Diet – Stable Isotopes - Stable isotope data for the reference lakes support the patterns observed in the stomach contents. In OP_R, approximately 25% of the perch had a trophic position of 4 or greater, indicating a switch to piscivory in the population (Fig. 5). This is confirmed by the C and N data (Fig. 6a). The trophic position of DA_R perch never exceeded four (Fig. 5). Stable C and N data for DA_R indicate that perch remain benthivorous (less negative $\delta^{13}\text{C}$, and trophic positions (TP) between 3 and 4 for all age classes past 2⁺). However, the lack of 1⁺ perch made it difficult to make inferences about their early diet patterns.

Perch in intermediate lakes (VA_I and BO_I) had isotopic patterns similar to those in DA_R (Fig. 6b). Like the perch in DA_R, the TP of perch from intermediate lakes never exceeded four (Fig. 5 and 6b). Unlike the perch in either of the reference lakes, 1⁺ and 2⁺ perch in VA_I and BO_I had no consistent changes in TP with $\delta^{13}\text{C}$ signatures. There was considerable variability in the TP of 1⁺ and 2⁺ perch. For example, in VA_I, 1⁺ perch had a mean TP of 2.95 ± 0.14 SD and in BO_I, the mean TP of 1⁺ was 3.34 ± 0.12 , whereas in OP_R, the mean TP of 1⁺ perch was 3.45 ± 0.08 . Therefore, the isotopic signatures of 1⁺ and 2⁺ perch were not indicative of planktivorous feeding (low TP, lighter $\delta^{13}\text{C}$) that progress to benthic feeding.

The diet patterns indicated by the stable isotope data of perch in the polluted lakes (Fig. 5 and 6c) were different from those using the stomach content data. In OS_P, approximately 25% of the perch had TP ≥ 4 (Fig. 5). This suggests that 4⁺ perch

cannibalize younger perch, as the only fish in OS_P are perch. In DT_P, the majority of perch have the same TP (between 3 and 4) (Fig. 5). However, a small number of perch in DT_P have TP \geq 4 (Fig. 5 and 6c). The TP and $\delta^{13}\text{C}$ suggest that these fish were feeding on forage fish (e.g. spottail shiner) that feed on epiphytic algae. The Y.O.Y. perch from DT_P have a high TP given their age. However, the isotopic signature of Y.O.Y. fish can be a reflection of the parental signature and not a true reflection of Y.O.Y. feeding (Vander Zanden et al. 1998). The $\delta^{13}\text{C}$ of DT_P perch does indicate a greater degree of benthic feeding than their stomach contents would suggest.

Fish size – Perch from reference and one intermediate lake (BO_I) achieved a significantly larger maximum weight than perch from polluted lakes or VA_I (Fig. 7, one way ANOVA, $F_{5, 79} = 83.16$, $p < 0.0001$, followed by Tukey-Kramer multiple comparison test). Perch in reference lakes were approximately 3-6 times heavier than perch in polluted lakes.

Discussion

The flow of energy in unpolluted systems - Benthic invertebrates are important components of aquatic food webs (Vadeboncoeur et al. 2001). The array of behaviours within the benthic community ensures that nutrients are not permanently stored in sediments, but will flow to higher trophic levels through a variety of pathways (Covich et al. 1999). In addition to the well-established role that benthic invertebrates play as detritivores and predators, benthic invertebrates are also pivotal in the processing of allochthonous inputs (leaf litter), biofilm, benthic algae, and macrophytes. The taxonomic diversity of the benthic community is related to its functional diversity (i.e. the diversity of resources processed by the benthic community) by the various feeding

strategies among benthic taxa. For example, shredders break apart coarse particulate organic matter into smaller particles that are consumed by other organisms that gather or filter fine particulate organic matter (Covich et al. 1999). This conversion of benthic resources into invertebrate production is important to higher trophic levels by providing food for secondary and tertiary consumers. For example, fifteen to 40% of freshwater benthic invertebrate production is lost to predation by fish (Strayer 1986). Exposure to heavy metals has the potential to disrupt food webs by severely reducing the abundance, biomass and distribution of animals at the base of the food web.

Aquatic ecologists have generally grouped possible energy pathways in lakes into benthic and pelagic paths. Research into lake food webs has been dominated by studies on the pelagic pathway, implicitly treating the two as parallel, but separate (Vadeboncoeur et al. 2002). This neglects the fact that many fish take advantage of the resources in both pelagic and benthic pathways as they change their niche (and thus diets) with growth. By altering the taxonomic and size composition of the benthic invertebrate community, heavy metal contamination has the potential to weaken the benthic pathway, forcing omnivorous fish like perch to rely more on energetically less profitable prey items like zooplankton and smaller benthic invertebrates.

Altered diet patterns in fish - Although reduced taxonomic diversity of the benthic invertebrate community can be used as an indicator of perturbation, on its own, it says little about the consequences to the food web processes to which benthic invertebrates contribute. In Rouyn-Noranda, the polluted study lakes were generally less diverse at the level of order and family than reference or intermediate lakes. The taxa that were lost or

greatly reduced from these systems are those that are generally considered sensitive to metal pollution (e.g. gastropods, ephemeropterans, and odonates).

The diversity and the range of biomass within the benthic invertebrate community can also explain changes in the diet patterns of yellow perch in the study lakes. In OP_R , all the diet data are consistent with perch that progress through three different diet stages. The percent occurrence data show that perch switch from planktivory after 1^+ and then switch to piscivory at 4^+ . The stable isotope data support this pattern with increases in trophic position at similar ages. The diet data for DA_R perch reflect fish that feed in an environment with abundant and diverse benthic resources. The percent occurrence data and isotopic evidence both suggest that perch do not switch to piscivory but remain benthivorous, although they do switch to larger prey items like crayfish and odonates. The perch in BO_I had similar diets as the perch in DA_R despite exposure to moderate levels of heavy metals. There is no indication from the percent occurrence data or the stable isotope evidence that BO_I perch switch to piscivory and like perch in DA_R , there is high prey taxa richness in their stomach contents. A large proportion of the older age classes fed on large ephemeropterans (*Hexagenia*), odonates and crayfish.

The patterns of diet in the perch from the reference lakes and BO_I contrast strongly with those from VA_I and the polluted lakes. In VA_I , there was relatively low prey taxa richness in the stomach contents. However, it is unlikely that VA_I perch show reduced taxonomic richness in their diet for the same reason that OS_P perch do. A large proportion of VA_I perch were consuming *Hexagenia* at all age classes. *Hexagenia* is an ephemeropteran that perch will consume in high quantities when *Hexagenia* are abundant in a system (Schaeffer et al. 2000). Therefore, the reduced taxonomic richness of the VA_I

perch diet is not necessarily due to reduced benthic diversity, but perhaps, a reflection of the availability of a preferred prey item that becomes large and abundant.

Perch from the polluted lakes (OS_P and DT_P) had very different diet patterns from those in the reference lakes and from each other. A large proportion of DT_P perch were planktivorous at all age classes, a pattern that is different from normal perch life history. In addition, the isotope data reflects the stomach content data in the reference and intermediate lakes. This was not true for the polluted lakes. The discrepancy between the patterns observed in the stomach contents and isotopes of the perch in polluted lakes is probably due to stronger seasonal fluctuations in their diets. In OS_P, piscivory has not been observed in the stomach contents in the year of this study or from early summer in previous years (Sherwood et al. 2000). However, the isotope data indicate that older perch are cannibalizing 1⁺ perch in OS_P. Therefore, in OS_P, piscivory occurs, but is not sustained in older perch. On the other hand, DT_P perch seem to be returning each spring to a juvenile feeding stage (i.e. planktivory). According to stomach content analysis, a large proportion of older perch (>3⁺) in DT_P still feed on zooplankton. However, the stable isotope evidence indicates that DT_P perch do rely on benthic invertebrates, and to a lesser degree, fish, for food. Therefore, early in the season, all age classes of DT_P perch rely on zooplankton but later in the season they seem to switch to benthivory, and a few switch to piscivory. In general, perch do not normally switch back to their previous diet stage once they are older than 2⁺ and have progressed to a different diet stage. Either they remain benthivorous, or they switch to larger, more energetically profitable prey. The presence of suckers (*Catostomus spp.*) in DT_P may partially explain the disrupted diet patterns in DT_P perch. Suckers can out compete perch for access to benthic

resources. Hayes et al. (1992) removed all suckers from a non-contaminated study lake and found that before treatment, adult perch stomach contents were dominated by zooplankton and, to a lesser degree, chironomids and small ephemeropterans. After removal of the suckers, the perch had switched to a primarily benthic diet, had increased mean weight of stomach contents and increased total body length. It is therefore plausible that in DT_P, the perch have to compete with suckers for access to a depauperate benthic fauna. In early June, all age classes of DT_P perch will forage in the pelagic zone of the lake. As the summer progresses, water temperatures rise and macrophyte cover increases, which can also increase the abundance of benthic invertebrates, reducing competition between suckers and perch. Furthermore, some perch in DT_P (i.e. perch with TP \geq 4) have found a niche where they can access prey fish. This, however, does not seem to be a general pattern for the perch in this lake.

The suitability of invertebrates as prey is partially related to their body size. The body size of individual prey items can alter the profitability of expending energy towards capture and consumption of that prey (Werner & Gilliam 1984, Sherwood et al. 2002). The polluted lakes have truncated size distributions relative to that of the reference lake (OP_R). The maximum size of individuals in OP_R was 13.5 mg d.w., whereas in OS_P and DT_P it was 3.5 mg d.w.. Mean prey size (excluding zooplankton) in DT_P and OS_P (0.80 ± 3.45 SE and 4.12 ± 3.08 SE respectively) indicate that perch in the polluted lakes are feeding within the available size range of the benthic community. In OP_R, the mean prey size (34.73 mg d.w.) greatly exceeds the size availability observed for its benthic community. Again, the increased amount of piscivory in OP_R can account for this difference. Overall, the average size of prey is significantly smaller in the polluted lakes

than reference or intermediate lakes. Furthermore, as perch get older in the reference and intermediate lakes, the size of prey in the stomach contents increases with age, whereas in the polluted lakes, there is almost no increase in prey size with age. Therefore, in the polluted lakes adult perch are expending energy foraging for less energetically profitable prey, which in turn has increased their activity costs (Sherwood et al. 2002).

Changes in benthic community and perch diet ultimately lead to stunted perch - Our data suggest that the various alterations in benthic communities (reduced taxonomic diversity and reduced size availability) can alter trophic interactions in metal-contaminated food webs. The growth performance of perch can be linked to these alterations. The growth patterns of the Rouyn-Noranda perch fall into the same two groups as suggested by the stomach content data – OP_R , DA_R and BO_I have similar maximum weights and OS_P , DT_P and VA_I have similar maximum weights. The perch from the first group attain normal maximum body sizes ($DA_R > OP_R \approx BO_I$) for perch, whereas the perch from the second group are stunted to various degrees (maximum size, $OS_P > VA_I > DT_P$). The perch from OS_P and DT_P exhibit stunted growth that would be expected from perch foraging in prey communities that have low diversity, low prey size availability, and high degree of site to site variability in prey abundance in uncontaminated environments (Heath & Roff 1996). It is, however, surprising that the two intermediate lakes would fall into different groups given that there are no significant differences in the bioaccumulation of metals in adult perch organs (Laflamme et al. 2000) and in invertebrate diversity. Prey sizes are larger in BO_I , but the difference between VA_I and BO_I is not significant. There is, however, a significant difference in the physiological response of the 1⁺ perch; VA_I 1⁺ had a

significantly greater degree of cortisol impairment (a standard measure of physiological stress) than BO_I perch (Gravel et al. 2002). This difference was not seen in adult perch (Brodeur et al. 1997, Laflamme et al. 2000).

Previous work on the bioenergetic performance of the perch in Rouyn-Noranda demonstrated that perch from OS_P, DT_P and VA_I do not differ significantly from the perch in OP_R or DA_R in their rates of prey consumption. They do differ in their conversion efficiencies (Sherwood et al. 2000) and in their activity costs (activity was not measured in VA_I or BO_I) (Sherwood et al. 2002)). The authors attributed the growth reduction observed in perch from polluted lakes to disruptions at the level of energy metabolism and not energy intake, as had been previously suggested by other studies (Munkittrick & Dixon 1988)) and that foraging efficiency was reduced in polluted systems, potentially due to simplified prey bases (Sherwood et al. 2002). Although Sherwood et al. (2000) did not directly measure invertebrate community abundance or size structure, the stomach content data of the perch showed similar differences in prey richness among lakes as the present study. This suggests that the patterns observed in the present study are consistent from year to year. Furthermore, we know that it is not a lack of total invertebrate biomass that can explain the effects seen at the level of growth – there were no significant differences between OS_P and OP_R, but lower levels of invertebrate diversity and size availability. It therefore seems that the shift towards a smaller, less diverse prey base has compromised the foraging efficiency of perch in polluted lakes. These increased foraging costs have contributed to the poor growth of these perch.

Although the stable isotope data indicate that some perch in OS_P and DT_P do become piscivorous, perch in either lake do not seem to benefit energetically from this diet change. In OS_P, the cannibalistic perch are significantly larger than the non-cannibal perch (i.e. TP < 4) (cannibal' mean body weight = 49.61 ± 3.01SE vs. non-cannibals' mean body weight = 30.00 ± 3.33SE, t-test, F_{1,38} = 19.15, p < 0.0001). However, the cannibals are still significantly smaller than the piscivorous perch from OP_R (TP ≥ 4, OP_R mean body weight = 115.77 ± 8.58SE, t-test, F_{1,28} = 45.59, p < 0.0001). Persson et al. (2000) postulated that energy gained from cannibalism in Eurasian perch from uncontaminated lakes is positively related to the body size of the victims and the time in the season that cannibalism occurs. Similarly, in DT_P, the few perch that according to their isotopic signatures do become piscivorous (mean body weight = 45.91 g ± 6.23 SE) are significantly larger than other adult perch (mean body weight = 21.07 g ± 2.17 SE) that remain benthivorous (t-test, F_{1,35} = 14.16, p = 0.0006). But the piscivorous perch in DT_P are still considerably smaller than the piscivorous perch in OP_R (t-test, F_{1,9} = 9.04, p = 0.015). Therefore, in OS_P and DT_P, perch do experience some energy gain from piscivory, but it is not sufficient to achieve relatively normal perch body sizes.

Indirect effects of metal exposure can be as important as direct toxicity - In polluted systems, it is clear that the physiologies of organisms can be impaired by the stress of coping with chronic exposure (Brodeur et al. 1997, Hontela et al. 1998, Laflamme et al. 2000). However, what is less clear is the contribution that indirect effects (i.e. effects mediated through the food web) have on animals at higher trophic levels. Although recent work by Beyers et al. (1999) and Sherwood et al. (2000, 2002) has made great

advances towards identifying the contributions of direct vs. indirect effects of exposure, there is still much that needs to be studied. Although direct toxic effects have been observed in perch from this region (Brodeur et al. 1997, Laflamme et al. 2000, Giguere 2002, Gravel 2002), alone, they cannot explain all of the changes observed in perch growth. The decrease in OS_P perch condition observed by Sherwood et al. (2000) between 1998 and 1999 cannot be explained by increased direct toxicity, as the level of metal exposure to the perch did not increase between those years. The more plausible explanation is that changes in the benthic community (i.e., dramatic drop in chironomid densities between those years, Sherwood et al. 2000) led to increases in foraging costs. Also, DT_P and OS_P both have similar levels of bioaccumulation of metals and induction of metallothionein (a detoxification protein) in perch liver (Laflamme et al. 2000), but significantly different degrees of stunted growth. The two lakes do seem to differ in the size availability of prey and the level of competition among fish taxa, which might explain the differences in growth.

Any perturbation to a food web that weakens or removes energy pathways will result in negative impacts at higher trophic levels. For example, Tyson & Knight (2001) have reported similar patterns for Lake Erie perch as those observed in this study. Before 1950, *Hexagenia*, trichopterans and dipterans dominated the benthic community of Lake Erie. After dramatic increases in phosphorus loading, there was a shift to smaller bodied benthic invertebrates (dominated by chironomids). When water quality was poor and the benthic community was simplified, dipterans and zooplankton dominated adult perch diet. As perch diet shifted, perch growth rates also decreased. The combined effects of water quality controls and the invasion of zebra mussels resulted in the return of larger

benthic invertebrates. Perch growth and recruitment responded positively to the presence of the larger benthos. Similarly, the invasion of non-native bass to lakes resulted in native lake trout shifting habitats and feeding on less energetically profitable prey (Vander Zanden et al. 1999a). This resulted in reduced growth rates and stunted fish (Pazzia 2000). Regardless of the mechanism, alterations in the energy flow among pathways have the potential to exert a strong negative impact on the energy gained by higher trophic levels.

To have a better understanding of the impacts of indirect toxic effect of metals, we need a better understanding of littoral benthic processes and their contributions to higher trophic levels. Studies such as this behooves us to further our understanding of food web interactions and the ecology of polluted systems if we are to be able to effectively prevent further degradation and predict impacts and trajectories of recovery.

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Table 1. Limnological characteristics and metal concentrations in the study lakes.

Note: The free metal ion concentrations and D.O.C. were measured using dialysers installed at 10 cm above the sediments and The Windermere Humic Acid Model; see Fortin and Campbell (1998) for details.

¹ data are from Giguere et al. *unpublished*.

² data are from Laflamme et al. 2000.

Lake	D.O.C. ¹	pH ¹	Total dissolved metals ¹		metals in sediments ²	
			Cd	Cu	Cd	Cu
<i>reference</i>	(mg C/L)		(nM)	(nM)	(nmol/g)	(nmol/g)
Opasatica (OP)	7.73	7.4	0.11	43.98	5.10	144.00
Dasserat (DA)	9.47	7.3	0.18	32.42	n.a.	n.a.
<i>intermediate</i>						
Bousquet (BO)	14.71	n.a.	1.06	56.05	10.00	143.00
Vaudray (VA)	9.03	7.10	0.81	54.25	12.00	238.00
<i>contaminated</i>						
Osisko (OS)	3.53	7.90	2.44	118.54	590.00	105 000
Dufault (DT)	5.53	7.00	7.72	254.24	200.00	19 400

Table 2. Abundance/m² of benthic invertebrate taxa, mean (\pm standard error of the mean) of benthic invertebrates in Rouyn-Noranda lakes. Numbers followed by the same letter are not significantly different (One-way ANOVA within taxa, among lakes, followed by a Tukey-Kramer multiple comparison test).

ORDER		OP	DA	BO	VA	OS	DT	p
amphipoda	mean	21.19 a	355.95 b	38.51 b	134.67 b	3.32 a	0.52 a	0.009
	s.e.m.	12.15	190.66	20.16	58.93	2.33	0.52	
coleoptera	mean	1.09 a	0.24 a	1.24 b	0.00 a	5.72 b	1.04 b	0.004
	s.e.m.	0.56	0.24	0.51	0.00	2.00	0.70	
diptera	mean	552.38 a	1388.14 a	326.10 b	511.23 a	659.32 a	261.04 b	0.018
	s.e.m.	160.95	447.28	98.39	146.91	178.27	54.18	
ephemeroptera	mean	493.80 a	100.27 a	54.85 a	60.03 a	0.33 a	39.34 a	0.481
	s.e.m.	377.10	64.09	15.38	20.58	0.33	23.52	
gastropoda	mean	335.94 a	479.74 a	220.65 a	240.69 a	0.49 b	0.52 b	0.004
	s.e.m.	78.64	174.84	57.01	79.34	0.36	0.52	
hemiptera	mean	5.95 a	0.48 a	3.42 a	0.85 a	0.33 a	0.00 a	0.034
	s.e.m.	2.65	0.48	1.18	0.85	0.33	0.00	
hydrachnidia	mean	25.71	92.63	10.87	25.78	18.19	11.21	<0.0001
	s.e.m.	8.18	19.91	3.71	7.35	7.63	5.24	
odonata	mean	9.88 a	8.75 a	6.21 a	15.14 a	4.09 a	0.00 a	0.39
	s.e.m.	4.58	6.74	2.17	8.04	2.63	0.00	
pelecypoda	mean	2493.72 a	1129.74 a	31.37 a	118.77 a	2491.69 a	37.27 a	0.65
	s.e.m.	1926.98	582.30	8.18	49.42	1734.90	28.65	
trichoptera	mean	51.86 a	147.00 b	28.57 a	52.49 b	70.28 b	19.93 a	0.006
	s.e.m.	19.17	28.55	5.53	18.07	27.98	8.07	

Lake	Size of benthic invertebrate (mg d.w.)		
OP	0.22	± 0.84	a
OS	0.16	± 0.30	a
DT	0.09	± 0.23	b

Fig. 1. Map of study area. This map shows the location, in relation to the copper smelting centre of Rouyn-Noranda, of 4 of the six lakes sampled in Abitibi, north-western Quebec (enlarged area, approximately 48°00"N, 79°00"W; average, relative wind directions at Rouyn-Noranda are indicated by vector diagram, modified from Couillard et al. 1995).

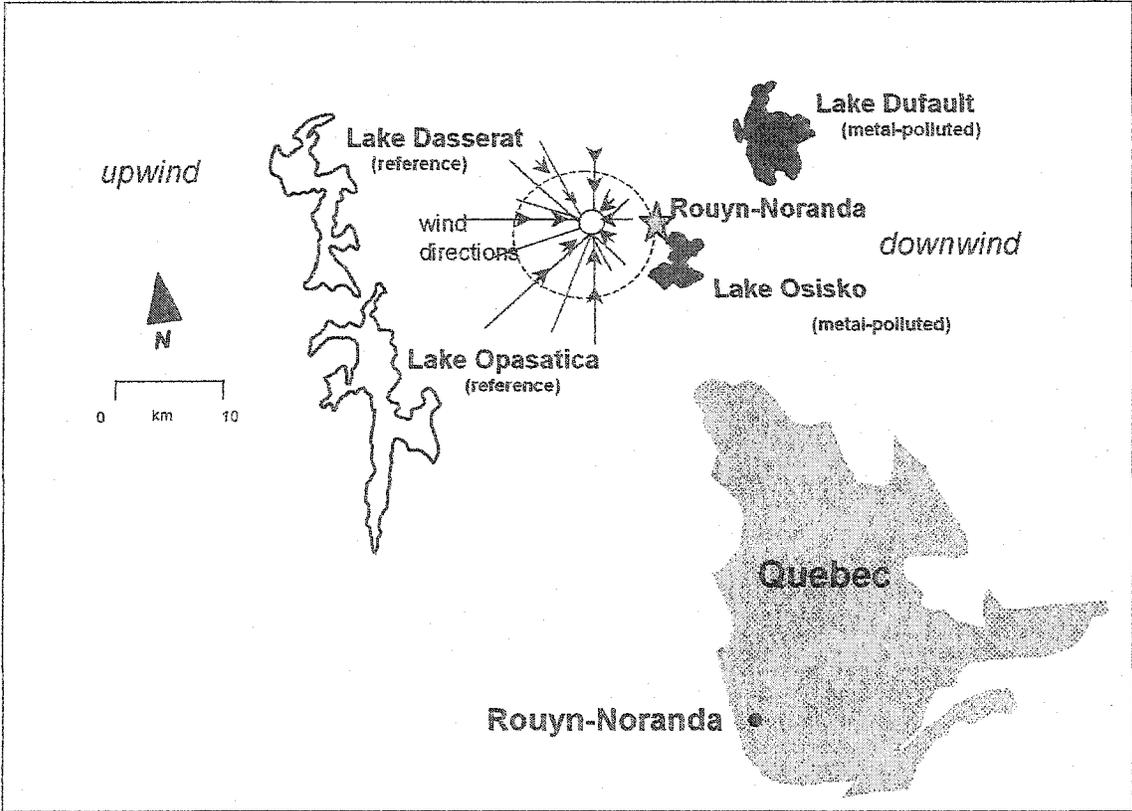


Fig. 2: Shannon-Weaver's diversity for each lake. Samples represent the mean diversity of taxa at the level of a) order and b) family. H' was calculated using the abundance of individuals within and among taxa. Lakes followed by the same letter are not statistically different from each other (one-way ANOVA)

Fig. 3. Mean weight of non-zooplankton stomach contents (mg dry weight), excluding 1+ perch, (mean \pm standard error), DA n = 31, OP n = 25 , BO n = 27, VA n = 16, OS n = 42, DT n = 38. Lakes followed by the same letter are not significantly different from each other (One-way ANOVA, $p = 0.05$, followed by a Tukey-Kramer multiple comparison test).

Fig. 4. Ontogenetic changes in the mean size of non-zooplankton prey in Rouyn-Noranda lakes. Perch older than 5+ were grouped into one age class. OP: prey size = $20.23 \text{ age} - 20.9$, DA: prey size = $18.38 \text{ age} + 4.52$, BO: prey size = $7.27 \text{ age} + 0.51$, VA: $7.05 \text{ age} + 2.83$, OS: prey = $1.68 \text{ age} + 0.23$, DT: prey size = $-0.12 \text{ age} + 4.16$ (one outlier removed).

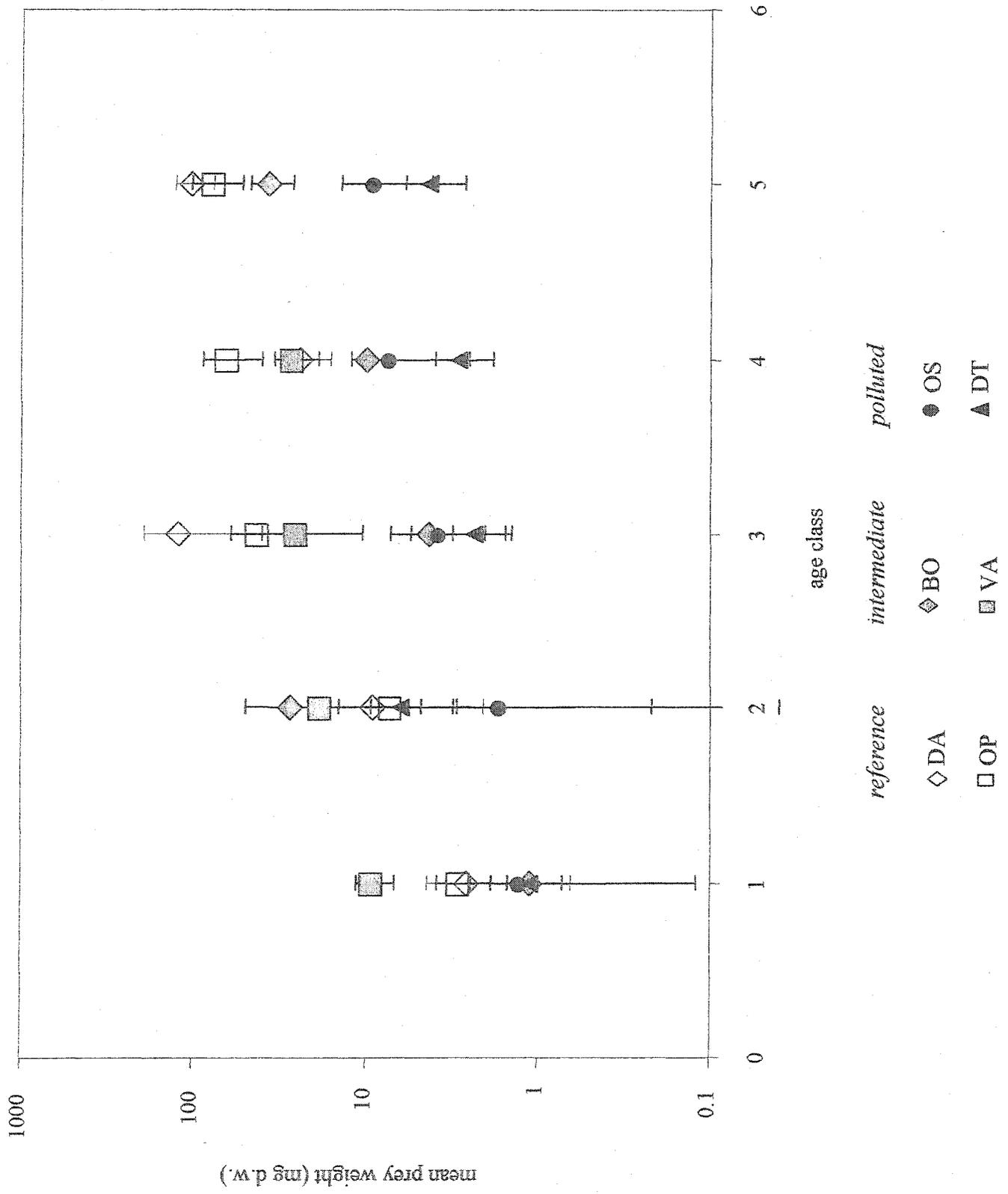


Fig. 5. Trophic position for all age classes of perch. A trophic position of 4 is indicative of piscivory. DA n = 30, OP n = 32, BO n = 23, VA n = 28, OS n = 61, DT n = 58.

Fig. 6. Trophic position of perch relative to the ^{13}C signature of perch (dark grey circles) and their prey items, zooplankton (clear circles), benthos (light grey circles), and minnows (clear circle with x).

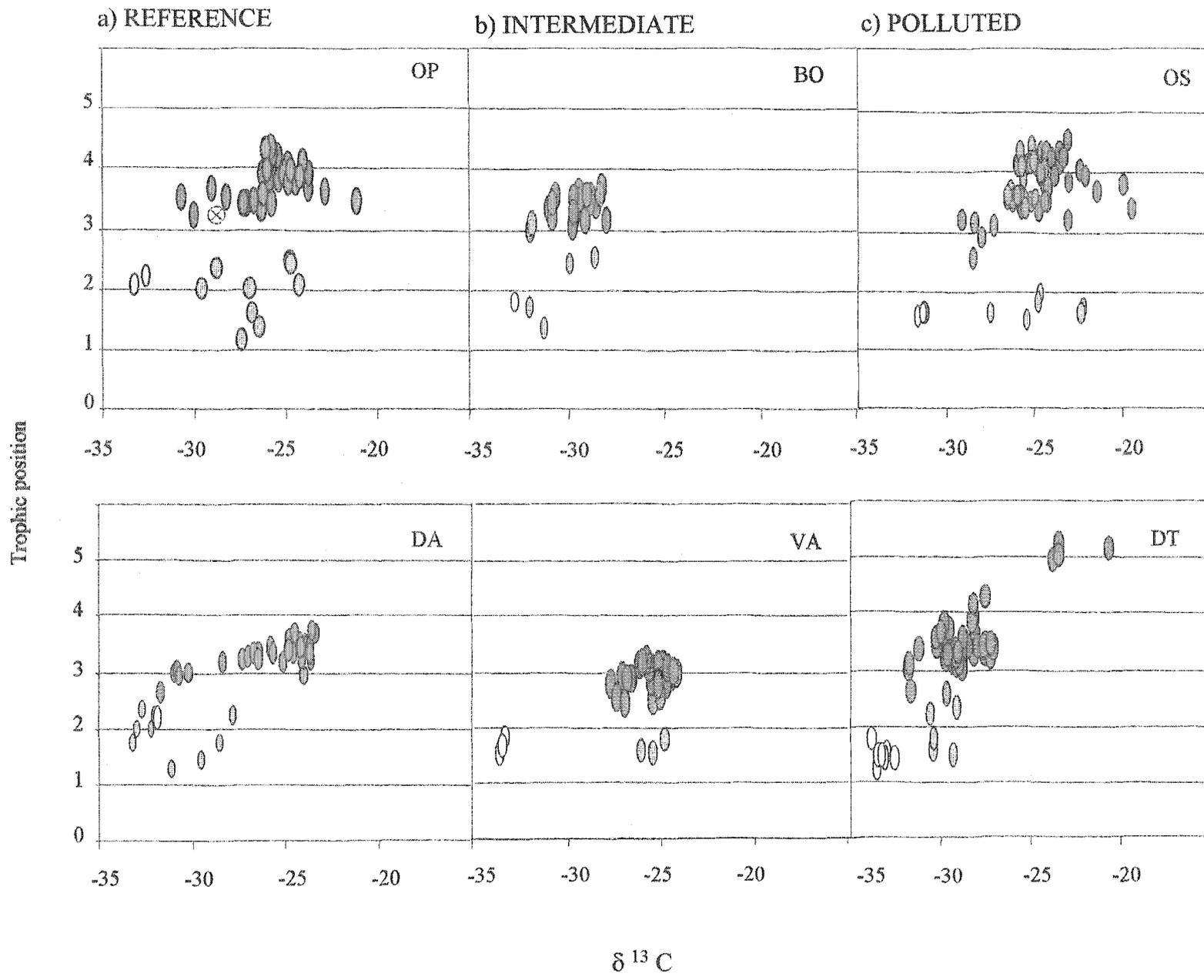
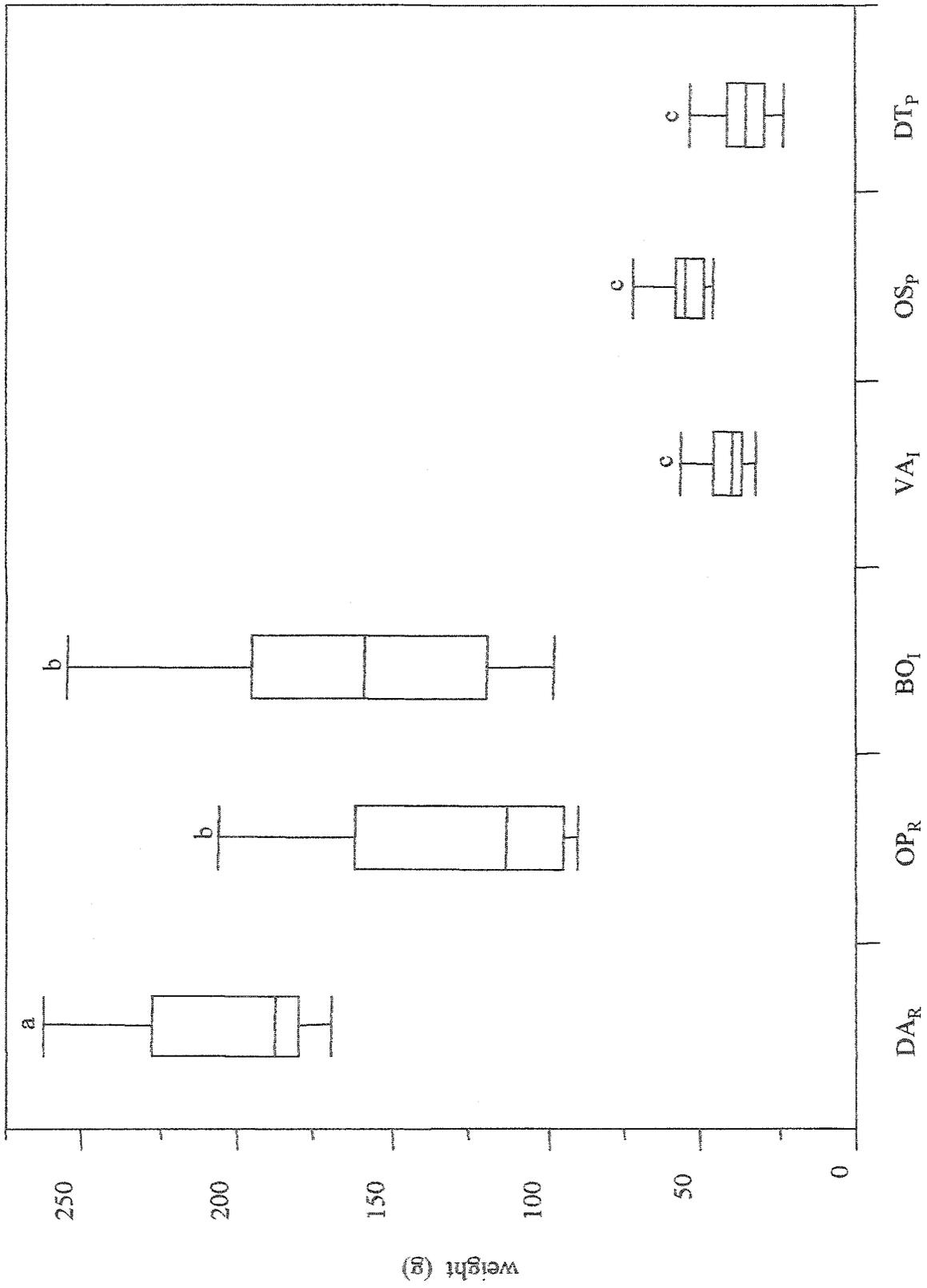


Fig. 7. The distributions of body weights for the largest 25% of perch among study lakes. Lakes with the same letter are not statistically significant from each other (One way ANOVA followed by Tukey Kramer multiple comparison test on the mean weight of perch in the largest 25% of the population.).



CHAPTER 2: The influence of ontogenetic diet shifts on metal exposure in yellow perch (*Perca flavescens*).

Introduction

In chronically polluted aquatic ecosystems, fish can be exposed to heavy metals through two principle routes of exposure: through contaminated food and from water, across their gills (Miller et al. 1993, Langston & Spence 1995, Szebedinszky et al. 2001). Although fish may be exposed to metals from two routes, the majority of experimental studies of metal uptake in aquatic organisms are based on exposures to contaminated water (Langston & Spence 1995). However, recent laboratory (Woodward et al. 1995, Ni et al. 2000, Szebedinszky et al. 2001) and field (Dallinger & Kautzky 1985, Besser et al. 2001) work has demonstrated that the trophic transfer of metals is a significant contributor to the accumulation of metals in target organs like liver and kidney of fish.

Experiments that test dietary exposure of metals generally provide fish with food that is uniform in size and metal concentration. The food type and quality in these experiments does not change throughout the course of the experiment. Most experiments also control the size or age of fish studied, and experiments are not usually long enough to include important life history changes. Thus, these studies do not reflect how wild fish will experience food-borne metals. Wild fish must forage for prey that is of various sizes, types, and nutritional quality. Concentrations of metals in prey may not be similar among taxa and within taxa in a polluted environment. In short, wild fish have highly heterogeneous diets that are different from the prey lab fish receive. The heterogeneity of prey can have a strong bearing on the outcome of the population level patterns of metal accumulation in fish. For example, Farag et al. (1999) found that the taxonomic differences among prey and the forms of metals in metal contaminated invertebrates fed to juvenile trout increased the bioavailability of the metals to the trout.

The most frequent life history characteristic used to study the effects of metals in fish is growth (Langston & Spence 1995, Sherwood 2001). This ignores a major factor in the life history of many fish – ontogenetic diet shifts. Many freshwater fish change the dominant prey type in their diet as they grow to optimize energetic performance (Werner & Gilliam 1984, Mittelbach & Persson 1998). Having a wide array of prey types and sizes allows omnivorous fish to meet the energetic demands of growth and helps to increase the efficiency of their foraging (Werner & Gilliam 1984). Thus, many fish undergo a benthivorous feeding stage during their lifetime (Mittelbach & Persson 1998, Vadeboncoeur et al. 2002). Yellow perch (*Perca flavescens*), a freshwater fish common throughout much of North America, will change its niche as it grows from being a pelagic, zooplankton predator to a littoral, benthic invertebrate predator. Although benthivory offers important energetic advantages to fish, it can also expose fish to high levels of contamination. Sediments are often the largest reservoir of metals in aquatic ecosystems (Luoma 1983). As a result, benthic invertebrates also tend to have high concentrations of heavy metals (Hare 1992, Farag et al. 1998). Thus, benthic invertebrates can also form an important link in the trophic transfer of metals in freshwater systems (Dallinger & Kautzky 1985, Woodward et al. 1994, Farag et al. 1999).

A fish can accumulate metals from its diet at each stage of its life, but the concentrations of metals in the diet may not be the same at each stage. The metal concentrations of invertebrates will be related to the concentration of metals in their surrounding medium (Hare et al. 2001, Chang & Reinfelder 2002), their diet (Smock 1983a) and the assimilation efficiency of the metals associated with prey (Reinfelder et

al. 1998). The metal contribution that invertebrate prey will make to the total accumulation in fish will also be mitigated by the concentration and bioavailability of the metal associated with the prey. Recent laboratory experiments (Baines et al. 2002) have shown that pelagic food webs (algae-zooplankton-fish) are not very efficient at transferring metals to higher trophic levels. The authors experimentally derived assimilation efficiencies for metals and then applied those numbers to a kinetic model for trophic transfer of metals. The model results agreed well with the concentration of metals from the same fish species in the wild. The authors further suggested that benthic food webs in the same habitat were potentially more efficient at transferring metals (Baines et al. 2002). Other experimental work has shown that zooplankton metal elimination rates can be an order of magnitude higher than benthic invertebrates' elimination rates (Reinfelder et al. 1998). In contrast, many benthic invertebrates become tolerant to metals in chronically polluted systems by sequestering and storing high concentrations of metals in their tissues (Hare 1992). Therefore, if fish change their niche from feeding in the pelagic on zooplankton to feeding on benthic invertebrates in the littoral zone, they risk increasing their exposure to metals. This pattern is more likely to occur in systems where the sediment concentrations of metals are high due to high historical loading or to natural enrichment of the catchment, and when the dissolved concentrations of metals are low. This scenario can result in zooplankton being at a lower metal concentration than benthic invertebrates. Increases in metal accumulation with ontogenetic diet shifts have previously been reported for marine turtles (Sakai et al. 2000) and benthic feeding stages in chronically exposed freshwater fish have been identified as a potential survivorship bottleneck (Amundsen et al. 1997, Farag et al. 1998,

Farag et al. 1999). However, no study has specifically tested that diet changes could act as a potential mechanism for changes in metal exposure.

Our objectives were to assess whether fish would accumulate more metals after switching from a diet of lower metal concentration (zooplankton) to one of higher metal concentration (benthic invertebrates). Specifically, we test the hypothesis that the concentration of Cu and Cd in the livers of perch will be positively correlated to the proportion of benthic invertebrates in the diet.

Methods

Study area – Fish and invertebrates were sampled from six lakes in the mining region of Rouyn-Noranda, north-western Quebec, Canada (48°00'N, 79°00'W). Lakes in this region are affected by former mining operations, current smelting activity, and atmospheric deposition. As a result, heavy metals have accumulated in the water column and sediments of lakes downwind from the city's copper refinery. The study lakes were chosen to represent a gradient of Cu and Cd contamination and a narrow range in limnological characteristics (Table 1). Two lakes upwind from the refinery, Opasatica (OP_R) and Dasserat (DA_R), were chosen as a reference sites. Lakes Osisko (south basin) (OS_P) and Dufault (DT_P) were chosen as high pollution sites. Although both DT_P and OS_P are considered high pollution sites, they differ in which abiotic compartment has the greatest concentration of metals (Table 1). Lakes Vaudray (VA_I) and Bousquet (BO_I), which lie several km downwind from the refinery, were classified as lakes with intermediate levels of pollution.

Fish sampling & invertebrate sampling – We sampled yellow perch (*Perca flavescens*) from the six lakes in June 2000 in the evenings. Methods of capture included gill nets and angling. We collected young of the year (Y.O.Y.) using a Mysis net that was towed within and just outside of the littoral zones of the lakes. Approximately 30-100 fish were selected representing as wide a range of fish sizes (2 – 30 cm) as possible. Perch were put on ice in the field, and immediately frozen upon return to the lab. Benthic invertebrates were collected from the littoral zones of the study lakes using a kicknet and an Eckman grab sampler. Zooplankton was collected by conducting horizontal and vertical tows in the epilimnion with zooplankton nets (mesh size: 250 µm). Invertebrates were placed in polyethylene bottles, placed on ice in coolers and returned to the lab. Benthic invertebrates were sorted and identified to family using published keys (Peckarsky 1990), rinsed with milli-Q-water, frozen, and freeze dried.

Perch Diet Analysis – We established the diet of perch by stomach content analysis and stable isotope analysis. Perch were thawed, and the opercula, stomach, and a small piece of dorsal white muscle were removed. Opercula were used to identify age class of perch. Diet diversity was estimated as percent occurrence, defined as the percentage of fish/age class/lake with a particular prey item in their stomach contents (Hyslop 1980, Marshall & Elliott 1997). We used stable isotopes to establish a more integrated measure of diet patterns. Stable ^{13}C signatures of consumers generally reflect the ^{13}C signature of their prey. Therefore, ^{13}C signatures are conserved from the bottom of the food web to the top (Deniro & Epstein 1978, Vander Zanden & Rasmussen 1999) and can provide information about the original source of carbon within a food web. The ^{15}N signature of

the consumers can be used as an indicator of trophic position within a food web because consumers become enriched in ^{15}N relative to their prey by ~3-4‰ (mean 3.4‰) (Deniro & Epstein 1981).

Proportion of benthivory – We used the following simple mixing model to estimate the proportion of benthic feeding in the perch:

$$(1) \quad \% \text{ Benthivory} = (\delta^{13}\text{C}_{\text{consumer}} - \delta^{13}\text{C}_{\text{zoop}} / \delta^{13}\text{C}_{\text{benthos}} - \delta^{13}\text{C}_{\text{zoop}}) * 100$$

This model differentiates the $\delta^{13}\text{C}_{\text{consumer}}$ from that of two endpoints, the average zooplankton signature and the average littoral benthic invertebrate signature. In some cases, the invertebrates chosen as endpoints could not adequately differentiate the isotopic signature of the consumer. These perch were eliminated from the % benthivory statistical analysis. However, these points were included in the correlations for $\delta^{13}\text{C}_{\text{consumer}}$ vs. the metal concentrations. The percent benthivory for each perch was arc sine transformed. Diets were considered fully switched to a new diet stage when $\geq 50\%$ of the diet was the new prey type (Hjelm et al. 2000).

It is generally accepted that inorganic metals do not biomagnify up the food web (Dallinger et al. 1987). We used the $\delta^{15}\text{N}$ of the perch to discriminate the trophic position of the fish. Trophic position was calculated as:

$$(2) \quad \text{TP} = ([\delta^{15}\text{N}_{\text{consumer}} - \delta^{15}\text{N}_{\text{baseline}}] / 3.4) + 2$$

Whole samples of invertebrate, pooled zooplankton and perch muscle tissue were frozen, dried at 80° C for 48 hours in a drying oven and ground into a fine powder using a mortar and pestle. Isotopic content was measured at the G.G. Hatch Laboratories, University of Ottawa.

Metal analysis: Perch livers were analysed individually when the weight of the liver exceeded 5-10 μg dry weight. Smaller livers were pooled until the weight reached 5-10 μg dry weight. The livers of perch with similar total body size ($\pm 0.5\text{cm}$) and gut contents were pooled. Invertebrates of the same taxa and similar body size ($\pm 0.1\text{cm}$) were pooled until a weight of 5-10 μg dry weight. Total length of invertebrates was measured using a dissecting microscope with an ocular micrometer. We chose invertebrate taxa for metal analysis based on their dominance in the diets of perch (Table 2). Perch livers and invertebrates were rinsed with milli-Q water to remove particles (in the case of the invertebrates), weighed in acid washed polyethylene containers and then either dried in a drying oven at 60°C , or frozen and then lyophilized.

Perch livers were digested following the methods of Borg et al. (1981). All lab utensils and containers were soaked in 15% solution of trace metal clean HCl & HNO₃ overnight and rinsed 3 times in milli-Q water. Individual liver samples were placed in pre-weighed tubes, and weighed. Ten ml of trace metal clean HNO₃ (Fisher) was added to each tube. The tubes were then placed in heating blocks in a fume hood and were heated to approximately 50°C for about 2h. After initial foaming ceased, the temperature was increased gradually to 110°C for 15-18 hr. Samples were evaporated to near dryness and then cooled. Trace metal clean H₂O₂ (30% solution) was added and the solution was then heated to 110°C for 5-6 hr, at which point fat residues would be oxidized. The solution was cooled again and a small amount of milli-Q water was added. If the solution was not clear, uncoloured and free of fat residues, more nitric acid was added again and evaporated. When the solution was clear and uncoloured, it was diluted to 5- 6 mL with

milli-Q water. Samples were analysed for Cd and Cu on a flame atomic absorption spectrometer (PE 3100). Invertebrate samples were sent to I.N.R.S.-Eau (Université de Québec, Sainte-Foy, Quebec, Canada) for analysis. These samples were analysed on an I.C.P. mass spectrometer.

Quality control- For every twelve tissue digestions, three laboratory reagent blanks and 2-3 certified standard reference materials were treated in the same manner as the tissue samples. The certified reference material for fish liver was DOLT-2 (National Research Council of Canada; certified value: Cu = 25.8 (± 0.5), Cd = 20.8 (± 1.1), numbers in brackets represent 95% confidence intervals). Measured values for DOLT-2 were: Cu = 26.55 (± 0.59) and Cd = 20.29 (± 0.49) (values in brackets represent \pm standard error). Standards containing Cd and Cu were analysed with every batch of metal analysis. Sample preparation for standards was done in a trace metal clean flow hood. All samples, certified reference material, and standards were measured in triplicate, with coefficients of variation for Cu and Cd less than 20%, and 75% of samples had coefficients of variation less than 10%. Minimum detection limits (MDL) for perch livers were: Cu = 0.09 mg/L and Cd = 0.02 mg/L. Certified reference material for invertebrates was TORT-2 (National Research Council of Canada; certified value: Cu = 106 (± 10.0), Cd = 26.7 (± 0.6), numbers in brackets represent 95% confidence intervals). Measured values of TORT-2 were: Cu = 104.9 (± 1.70) and Cd = 27.4 (± 0.20). MDL for invertebrates was Cu = 0.5 $\mu\text{g/g}$ and Cd = 0.3 $\mu\text{g/g}$. Samples below the MDL were substituted with MDL/2. This substitution causes a slight upward bias in the statistical analysis (AllenGil et al. 1997).

We used the total length of perch, the proportion of benthivory, and the concentration of Cd and Cu in perch livers as variables in a Pearson correlation matrix. Correlations were run with and without perch that were identified as piscivorous by their trophic position ($TP \geq 4$).

Results

Metal levels in invertebrates and zooplankton – In the reference lake OP_R, the concentrations of metals in zooplankton were very similar to the average concentrations in benthic invertebrates (Cu: 11.30 $\mu\text{g/g}$ d.w. vs. 14.9 $\mu\text{g/g}$ d.w., respectively) and slightly lower than in the primary diet item (Table 3). In DA_R, the concentrations of metals in the zooplankton were 4 to 20 times lower than the primary diet item metal concentration (Table 3). In the intermediate lakes (VA_I and BO_I), the Cu concentrations in the zooplankton were similar (average benthic invertebrates were 1 to 1.5 times greater than the zooplankton concentrations) (Table 3). In contrast, in BO_I, the Cu concentration of the primary diet item was 3 times lower than the zooplankton. There was approximately 13 to 27 times more Cd in the average benthic invertebrates than the zooplankton for both intermediate lakes. In OS_P, the Cu concentrations of the primary diet item was 18 times greater than the zooplankton and Cd concentrations were 2.5 times greater in the primary diet item. In DT_P, the Cu concentration of the average benthic invertebrate was approximately twice the concentration of the zooplankton, but the zooplankton was approximately the same concentration as the primary diet item. The Cd concentration was higher in the zooplankton than in the primary diet item (Table 3).

Metal concentrations in perch livers – On average, the Cu concentrations in the livers of perch were significantly higher in the two polluted lakes (OS_P and DT_P) (one-way ANOVA and Tukey-Kramer test $F_{5, 195} = 18.5$, $p < 0.0001$), whereas, there was no significant difference in the concentration of Cu in the livers of OS_P and DT_P perch (Table 4). The Cd concentrations in perch livers from the polluted lakes and one of the intermediate lakes (VA_I) was significantly higher than the two reference lakes and the other intermediate lake (BO_I) (Table 4) (one-way ANOVA, and Tukey Kramer test $F_{5, 189} = 21.65$, $p < 0.0001$). Mean DT_P perch liver [Cd] was significantly higher than OS_P and VA_I (Table 4).

Predicted relationship between metals in the liver and diet – Given the small differences between the concentrations of metals in the zooplankton and the primary diet items, we expected that there would be no significant correlation between the proportion of benthivory and metal accumulation in the liver of perch from OP_R in either Cu or Cd. For perch from DA_R, we expected a significant positive correlation between the proportion of benthivory and liver metal concentrations for both metals (Table 5). In the intermediate lakes, we did not expect any significant correlations between the proportion of benthivory and liver Cu concentration, but we did expect a significant positive relationship for Cd (Table 5). In OS_P perch, we expected a significant, positive correlation between benthivory and concentration of both metals in the liver. In DT_P, we did not expect any significant correlations for either metal (Table 5).

Observed relationship between liver metal concentration and diet - The two reference lakes had different patterns in the correlates with liver metal concentrations. In OP_R, there were no significant correlations between fish length, proportion of benthivory and the concentrations of metals in the liver of perch, except for a negative correlation between Cu and the percent benthivory. In DA_R, total length of perch was significantly correlated to the proportion of benthivory and the concentration of Cu and Cd in the liver. However, the percent benthivory was a better correlate to the metal concentrations in perch livers (Table 5). In both VA_I and BO_I, there were no significant correlations between any of the correlates except for total length of perch and Cd liver concentrations (Table 5). In OS_P, benthivory was an equally good correlate as length for the concentration of Cu in perch livers (Table 5). Although the proportion of benthivory was significantly correlated to Cd concentrations in OS_P, total length was a better correlate (Table 5). In DT_P, length was the only variable that was significantly correlated to the concentration of metals in perch liver (Table 5).

Discussion

Understanding the various factors that contribute to the exposure and accumulation of metals in biota is important for developing appropriate and effective management tools. Researchers have made progress in understanding the complex interactions that occur between fish gills and metal laden water that surrounds them (Playle 1998, Hollis et al. 2000, McGeer et al. 2000). The role dietary uptake plays in metal accumulation is less clear. We know that the amount of metals that accumulate from the diet is mitigated by several factors, among them are: 1) the metal concentration of the prey, 2) the metal bioavailability in the prey, 3) the assimilation efficiency of

metals in prey by predators, 4) the ingestion rate of the prey by the predator's diet. As a fish grows and changes its diet, the magnitude and importance of each one of these factors may be different. For example, gut passage time is longer in larger, older fish because intestines are longer, and as a result, assimilation efficiencies can be higher for older fish (Baines et al. 2002).

The concentrations of Cu and Cd in the zooplankton and benthic invertebrates of lakes in Rouyn-Noranda were generally a reflection of the concentration of metals in the surrounding media. Zooplankton had higher concentrations of Cd and Cu in lakes with higher dissolved concentrations of these two metals (Fig. 1). Benthic invertebrates had higher concentrations of metals in lakes with higher sediment concentrations of Cu and Cd (Fig. 2). We did not quantify the assimilation efficiency or bioavailability of the metals from the prey to perch, but we did quantify the relative contributions that the prey made to the diet of the perch (Table 2). Feeding ecology of perch in the polluted lakes has been disrupted by exposure to metals. In all the lakes, a high proportion of the stomach contents consisted of benthic invertebrates. However, in the polluted lakes, the diets of adult perch were dominated by chironomids and zooplankton (Table 2). DT_P perch were consuming relatively high proportions of zooplankton at all age classes, a pattern not seen in perch from any of the other lakes. In perch from reference and intermediate lakes, there was greater diversity in the benthic invertebrates in the stomach contents, while zooplankton were only important to 1^+ perch. Interestingly, consumption rates in perch from polluted lakes were generally not significantly different from those in the reference lakes (Sherwood et al. 2000).

On average, perch from the polluted lakes (OS_P and DT_P) accumulated more Cu and Cd in their livers than perch from reference lakes (Table 4). However, given the differences in metal concentrations in the water column, sediment and prey, it is possible that different routes of exposure are important in each of the lakes. In lakes where the concentrations of both Cd and Cu were greater in benthic than in zooplanktonic prey (OS_P and DA_R), there were significant correlations between benthivory and metal accumulation (Table 6). In OS_P, the difference between the concentrations of metals in zooplankton vs. benthic invertebrates was the greatest among all the lakes. We therefore expected that OS_P perch should exhibit the strongest correlation between the proportion of benthivory in their diet and their metal accumulation. OS_P perch exhibited a positive correlation between the percent benthivory in their diet and metal accumulation in their livers. Furthermore, when piscivorous fish were removed from the analysis, the correlation between benthivory and metal accumulation increased to $r = .47$ (Cd) and $r = .53$ (Cu). However, the strongest correlations between benthivory and metal concentration were for DA_R perch (Table 6). The total accumulation of metals in DA_R perch livers was significantly lower than the perch from OS_P and were not at levels that elicited any negative physiological (Laflamme et al. 2000) or negative growth response (Laflamme et al. 2000). Despite the lack of toxicological response, these results do suggest that the accumulation of metals in DA_R perch was related to the diet of the perch.

In all other study lakes, the Cu concentrations in the zooplankton and benthic prey taxa were likely too similar to cause a change in uptake once the diet had switched from primarily zooplankton to primarily benthic invertebrates. This was also true for the Cd concentrations in the prey for OP_R and DT_P. We were therefore not expecting to observe

significant positive correlations between metal accumulation and percent benthivory in these lakes. Indeed, perch from OP_R, VA_I, BO_I, and DT_P did not demonstrate any significant positive correlations between the degree of benthivory and their accumulation of metals. In OP_R, we observed negative correlations with percent benthivory. In this lake, the perch begin feeding on fish at a younger age than perch in the other study lakes (Kövecses et al. *in prep*). This might explain the negative correlation with percent benthivory, as it would be expected that there would be less metals transferred from food at piscivorous stages.

It was somewhat surprising that there was no significant correlation between the proportion of benthivory and Cd concentrations in perch from the intermediate lakes, as the difference in Cd concentration in zooplankton was considerably lower than the benthic invertebrates. The lack of significant correlation may be due in part to differences in the bioavailability of Cd from benthic prey in the intermediate lakes. Bioavailability of metals from invertebrates to predators can be influenced by the partitioning of the metals between the surface and interior tissues in aquatic insects (Hare 1992). The proportion of metals partitioned internally vs. externally varies among taxa. In laboratory studies, when whole body Cd concentrations were approximately 10 µg/g in *Hexagenia*, approximately 40% of the Cd was bound to the chitinous body surface (Hare et al. 1991). In contrast, there was no significant accumulation of Cd on the outer surfaces of chironomids (Timmermans et al. 1992). In the intermediate lakes, ephemeropterans, especially *Hexagenia*, were among the most important prey items for perch. In contrast, in OS_P, chironomids were the only important prey in perch stomach contents. Thus, the taxonomic differences in the diet of perch may account for the lack of

expected pattern in the perch from the intermediate lakes. Farag et al. (1999) found similar results in an experimental study with juvenile salmonids. When juvenile trout were fed diets of wild invertebrates collected from various polluted streams, the invertebrate diet with lower metal concentrations resulted in higher metal burdens in trout. This heightened bioavailability was attributed to the forms of metals associated with the different taxa of each diet.

The absence of a positive correlation between benthivory and liver metal concentration in DT_P does not necessarily rule out dietary exposure as a significant contributor to total accumulation of metals. The shift in metal exposure with diet would not be identifiable in this lake because both zooplankton and benthic invertebrates are at relatively high metal concentrations. Furthermore, perch in DT_P had anomalous diet patterns – zooplankton forms a large proportion of the stomach contents of all age classes of perch in DT_P (100% of 1⁺ and 2⁺ perch to 43.75% of perch 5⁺ and older). Therefore, it would not be possible to identify a clear diet shift in the perch from this lake. Despite this, dietary uptake in DT_P cannot be fully discounted. Of all six lakes, DT_P perch had the highest concentrations of metals in the digestive tract. Furthermore, the concentrations of metals in the digestive tract were higher than in the gills (Giguère et al 2002). This suggests that metals are passing through the gut tissue in DT_P perch. However, DT_P has the highest dissolved concentration of metals, therefore, it is likely that both routes of exposure are important in this lake.

The large amount of variability in metal concentrations in the fish made it difficult to detect strong correlations. Other studies on perch from the same lakes have found similar degrees of variation in metal burdens in adult perch livers (Laflamme et al.

2000). There are several potential reasons for this variability. The concentration of metals in prey is not homogenous. Metal accumulation in invertebrates varies with individual size, feeding strategies and taxonomic groups (Smock 1983a, Smock 1983b). Bioavailability of metals may also be different among invertebrate taxa. Perch are opportunistic, omnivorous fish and will therefore have a high degree of prey diversity in their diet, both over time, within individuals and among individuals at any time. Therefore, within any given population of perch, there is the potential for considerable variability in the amount of metals to which they are exposed.

Patterns of metal trophic transfer may change with time in chronically polluted systems. It is a well-established pattern that chronic metal exposure creates predictable changes in the taxonomic composition of benthic invertebrate communities – metal sensitive taxa are eventually replaced with tolerant taxa. However, tolerant taxa have different mechanisms for withstanding the negative impacts of metals (Dallinger 1994). They either have storing mechanisms (e.g. metal binding proteins or metal containing granules) that allow them to accumulate metals (Seidman et al. 1986, Groenendijk et al. 1999) or they have various mechanisms for eliminating metals (Burgos & Rainbow 1998). Wallace et al. (1998) found that Cd associated with metallothionein in a deposit feeding oligochaete was more bioavailable to an omnivorous shrimp than the Cd bound to the oligochaete's granules. In this case, the prey detoxification mechanism mediated the trophic transfer of Cd. A fish whose diet is dominated by prey that use one or the other mechanism might either have the effect of food web transfer exacerbated or diminished (Dallinger et al. 1987). This hypothesis, however, has not yet been tested in wild fish.

Despite the moderate correlations found in this study, these results still provide compelling evidence that life history changes like ontogenetic diet changes may play an important role in the accumulation of metals in fish. Although several studies have suggested that benthic feeding may be important to metal accumulation in fish, no studies have linked variations in patterns of metal accumulation to diet shifts. Metals accumulated through the diet have already been linked to reduced growth (Lundebye et al. 1999), increased histological damage (Woodward et al. 1994) and decreased fecundity (Munkittrick & Dixon 1988) in adult and juvenile fish. Given that there are many fish species that undergo ontogenetic diet changes (Mittelbach & Persson 1998), our results support the idea that this life history characteristic is an important determinant in the potential for metals to accumulate in fish. Therefore, in order to establish effective risk assessment and regulations for protection of aquatic resources, all the life history factors that can contribute to exposure and accumulation of contaminants in fish must be taken into account.

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Table 1. Limnological characteristics and metal concentrations in the study lakes.

Note: The free metal ion concentrations and D.O.C. were measured using dialysers installed at 10 cm above the sediments and the Windermere Humic Acid Model; see Fortin and Campbell (1998) for details.

^a data are from Giguère et al. *in prep.*

^b data are from Laflamme et al. 2000.

Lake	D.O.C. ^a	pH ^a	Total dissolved metals ^a		metals in sediments ^a	
			Cd	Cu	Cd	Cu
<i>reference</i>	(mg C/L)		(nM)	(nM)	(nmol/g)	(nmol/g)
Opasatica (OP)	7.7	7.4	0.1	43.9	5.1	144.0
Dasserat (DA)	9.5	7.3	0.2	32.4	n.a.	n.a.
<i>intermediate</i>						
Bousquet (BO)	14.7	n.a.	1.1	56.1	10.0	143.0
Vaudray (VA)	9.0	7.1	0.8	54.3	12.0	238.0
<i>contaminated</i>						
Osisko (OS)	3.5	7.9	2.4	118.5	590.0	105 000
Dufault (DT)	5.5	7.0	7.7	254.2	200.0	19 400

Table 2. Important invertebrate prey items in the stomach contents of perch. Percent occurrence was calculated as the # of perch with a particular prey item in the stomach contents. Data are for all age classes of perch. OP, n = 55, DA = 55, VA = 39, BO = 40, DT, n = 76 and OS, n = 90.

^aData are from Kövecses et al. 2002, *in prep.*

	% occurrence ^a						Mean abundance of prey in stomach contents ^a					
	OP	DA	VA	BO	DT	OS	OP	DA	VA	BO	DT	OS
Amphipoda	3.6	52.8	6.3	40.0	0.0	0.0	0.1	5.2	0.1	4.7	0.0	0.0
Diptera	14.6	22.2	29.2	40.0	53.9	55.8	0.23	0.4	0.4	1.3	6.5	13.5
Ephemeroptera	14.6	11.1	43.7	70.0	0.0	0.0	0.4	0.1	1.7	2.4	0.0	0.0
Odonata	12.7	55.6	6.3	7.5	0.0	1.2	0.5	1.5	0.1	0.3	0.0	0.0
Tricoptera	7.3	33.3	2.1	22.5	11.1	12.8	0.4	0.7	0.0	1.5	0.6	0.4
Zooplankton	10.9	5.6	31.3	32.5	73.7	43.0	39.2	0.1	4.9	31.0	742.4	58.4

Table 3. Whole body metal concentrations for zooplankton and benthic invertebrates. Numbers in brackets represent one standard error. Primary diet item is the benthic invertebrate that was one of the most important in terms of its percent occurrence and the average abundance in the stomach contents (OP_R: odonates, DA_R: amphipods, BO_I & VA_I: *Hexagenia*, OS_P & DT_P: chironomids). All samples were a minimum of 5 – 10 µg d.w.

	<i>Reference</i>		<i>Intermediate</i>		<i>Polluted</i>	
	OP	DA	BO	VA	OS	DT
<i>Cu⁺ (µg/g d.w.)</i>						
zoop	11.30	2.96	17.21	9.48	37.18 ± (10.40)	47.55
avg benthic invertebrate	14.90 ± (4.90)	33.50 ± (12.65)	18.4 ± (6.70)	14.25 ± (2.64)	397.55 ± (132.13)	86.25 ± (50.75)
primary diet item	16.60	59.2	6	11.45 ± (1.05)	661.70 ± (117.103)	35.50
 <i>Cd⁺ (µg/g d.w.)</i>						
zoop	0.81	0.87	1.12	0.47	5.78 ± (2.09)	4.81
avg benthic invertebrate	1.17 ± (0.35)	5.69 ± (1.32)	14.1 ± (3.38)	11.15 ± (2.08)	11.92 ± (3.08)	10.10 ± (7.10)
primary diet item	0.60	3.4	16.7	13.26 ± (2.16)	13.43 ± (3.17)	3.00

Table 4. Concentration of metals in the livers of perch for the study lakes. Numbers are means for all age classes followed by ± 1 standard error. OP n = 33, DA n = 27, BO n = 21, VA n = 26, OS n = 53, DT n = 41. Numbers followed by the same letter are not significantly different, one-way ANOVA, and Tukey Kramer test $F_{5, 189} = 21.65$, $p < 0.0001$

	lake	n	liver [Cu] μg/g d.w.	liver [Cd] μg/g d.w.
<i>Reference</i>	OP	33	11.23 (±1.14) b	3.73 (±0.46) c
	DA	27	12.79 (±1.65) b	3.31 (±0.45) c
<i>Intermediate</i>	VA	26	15.99 (±1.03) b	16.30 (±1.94) b
	BO	21	25.19 (± 5.07) b	12.20 (±1.47) c
<i>Polluted</i>	OS	53	70.46 (± 9.03) a	25.05 (± 2.49) b
	DT	41	93.99 (± 12.13) a	35.58 (± 4.16) a

Table 5. Predicted direction of correlations between the proportion of benthivory in perch diet (asin ben), the concentration of Cd and Cu in perch liver (log liver [Cd or Cu], and total body length of perch. (/) = no significant correlation, (+) = significant positive correlation, (-) = significant negative correlation. Grey boxes indicate where our expectations deviated from the observed patterns. We expected to find significant positive correlations when the metal concentrations of benthic invertebrates exceeded the metal concentrations of zooplankton, a negative correlation when zooplankton were at a higher correlation than benthic invertebrates and no significant correlation when there was a negligible difference between zooplankton and benthic invertebrates.

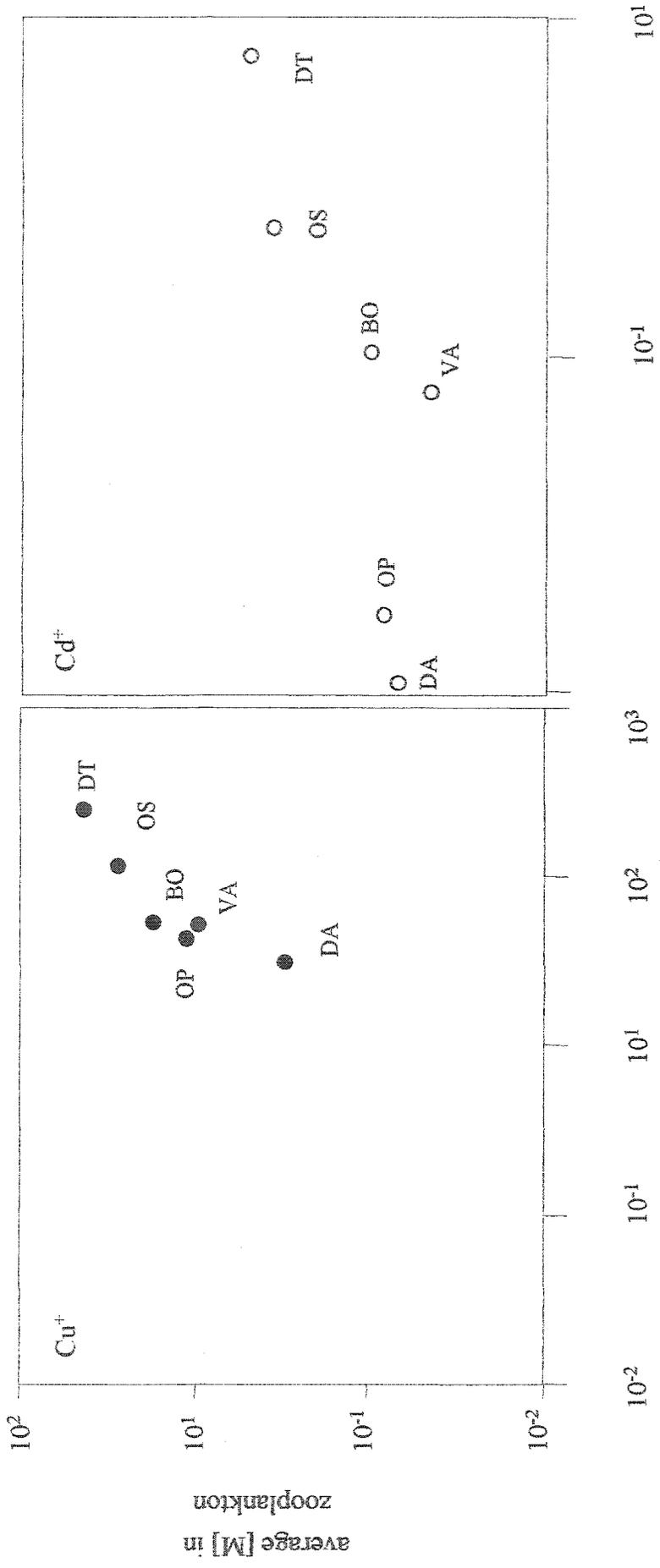
	OP		DA		BO		VA		OS		DT	
	pred.	obs.										
Cu	/	-	+	+	-	-	/	/	+	+	/	/
Cd	/	/	+	+	+	/	+	/	+	+	/	/

Table 6. Pearson's correlation for total length of perch (length), proportion of benthivory in the diet (asin ben), and the log transformed metal concentration of perch livers (log liver [M]). * = $p < 0.05$, ** = $p < .001$, *** = $p < 0.0001$, and n.s. = not significant.

		<i>Reference</i>		<i>Intermediate</i>		<i>Polluted</i>	
		OP	DA	VA	BO	DT	OS
asin ben	length	0.30 n.s.	0.83 ***	0.42 **	0.10 n.s.	0.40 **	0.59 ***
log liver [Cu]	length	-0.07 n.s.	0.53 **	0.22 n.s.	0.11 n.s.	0.39 **	0.48 ***
log liver [Cu]	asin ben	-0.63 ***	0.68 ***	-0.36 n.s.	-0.48 *	0.15 n.s.	0.48 **
log liver [Cd]	length	-0.05 n.s.	0.42 *	0.49 **	0.72 ***	0.60 ***	0.64 ***
log liver [Cd]	asin ben	-0.25 n.s.	0.49 *	0.19 n.s.	-0.43 n.s.	0.00 n.s.	0.36 **

Fig. 1. The relationship between the concentration of metals dissolved in the water column and the average concentration of metals in zooplankton. Note the different scales for the x-axis.

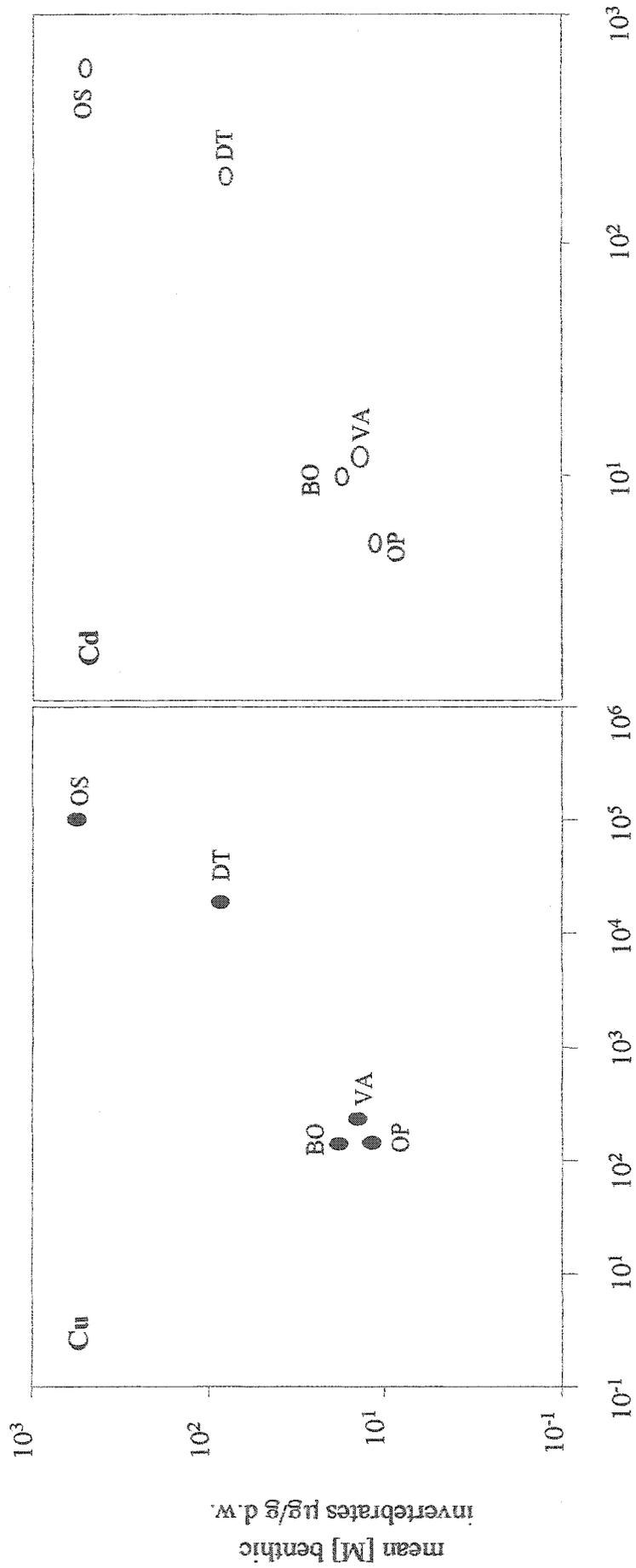
^a dissolved metal data are from Giguère et al. *in prep.*



total dissolved [M] nmol/L

Fig. 2. The relationship between the concentration of metals in the sediments of lakes in Rouyn-Noranda and the mean metal concentration in benthic invertebrates.

^b Sediment metal data are from Laflamme et al. 2000.



sediment {M} nmol/g d.w.

GENERAL CONCLUSIONS

Food web interactions can provide insight into the impacts of heavy metal contamination in aquatic ecosystems (Sherwood et al. 2002). Despite this, the subject has received little attention in the ecotoxicology literature (Carlisle 2000). However, a few previous studies have shown that the indirect effects of contamination (i.e. those mediated through the food web) can account for decreased growth and fecundity in adult white suckers (Munkittrick & Dixon 1988, Munkittrick et al. 1991) and decreased conversion efficiencies (Sherwood et al. 2000) and increased activity costs (Sherwood et al. 2002) in yellow perch.

In this thesis, I found that the loss of taxonomic diversity in the benthic invertebrate communities of metal contaminated lakes resulted in a shift towards smaller mean body size of benthic invertebrate. The changes in the benthic communities had a two-fold effect on the feeding ecology of perch: 1) perch diet shifted from a diverse diet consisting of a wide range of prey taxa to a diet dominated by chironomids or dominated by zooplankton and chironomids and 2) perch in contaminated lakes had significantly smaller mean prey size (non-zooplankton prey) than perch from reference lakes. Other aspects of the feeding ecology of perch was disrupted by metal contamination as well. For example, the isotopic signatures of perch from OS_P were indicative of cannibalism, a characteristic not seen in any of the perch from reference lakes. Perch from DT_P had an incomplete diet shift from zooplankton to benthivory – a high proportion of perch at all age classes were still feeding on zooplankton, a pattern that is not typical for perch from reference sites.

The shift towards less energetically profitable prey in the contaminated lakes has, at least partially, contributed to poor growth in the perch. In the contaminated lakes, adult perch showed high degrees of stunting. These results support previous findings which demonstrate that perch from the same polluted lakes have higher foraging costs (Sherwood et al. 2002).

This thesis also helps to highlight the idea that any perturbation (e.g. metal exposure, eutrophication, or even perhaps, acidification) that weakens benthic processes, can result in the same type of effects on higher trophic levels. Benthic and pelagic habitats are often treated as discrete food webs (Vadeboncoeur et al. 2002), with the implication that organisms rely only on one of the two main pathways for resources. However, it is well known that many fish shift their habitat with growth (Mittelbach & Persson 1998) and therefore, over the course of their lifetime, depend on both pathways for their energy. Thus, by diminishing or removing access to benthic energy sources, human induced perturbations can eliminate the benthic pathway as an energetically viable alternative energy pathway.

In addition to the various energetic benefits of maintaining benthic pathways, there is the potential for negative side-effects. We found that metal accumulation in perch livers was moderately correlated to the proportion of benthivory in the diet of perch. This was true in lakes where the concentration of metals was considerably higher in the benthic prey than the zooplankton prey. Although much work has been done to clarify the mechanisms of metal uptake across fish gills (Playle 1998, McGeer et al. 2000), there is less information on how diet can effect metal uptake in fish. The results

from the second chapter show that feeding ecology merits further study if we are to be able to accurately predict metal uptake in fish.

Studies such as these demonstrate that the effects of metal contamination are more complicated than what would be predicted simply by standard concentration dependent exposure – inter and intraspecific interactions, feeding ecology and ontogenetic life history changes all play a part in determining the effects of metal exposure in aquatic ecosystems.

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APPENDICES

APPENDIX 1
Perch life history data.

OSISKO (OS)

FISH #	TOT L (cm)	age	WT (g)	d13C	d15N	TP litt	%ben
1	16.40	6	39.35	-25.49	17.40	4.04	0.70
2	19.20	6	59.56	-24.57	18.34	4.32	0.81
3	16.80	7	48.20	-25.01	17.96	4.21	0.76
4	17.60	7	56.52	-23.53	17.62	4.11	0.94
5	17.90	4	49.77	-25.92	17.64	4.12	0.65
6	15.40	5	37.18	-24.41	16.52	3.78	0.83
7	15.60	5	32.69	-25.12	17.90	4.19	0.75
8	15.80	4	40.00	-25.32	17.65	4.12	0.72
9	18.00	7	55.58				
10	17.60	6	53.87				
11	19.40	5	68.70				
12	16.70	6	45.09	-25.15	17.62	4.11	0.74
13	16.50	6	35.27	-25.62	16.80	3.87	0.69
14	16.60	5	45.40				
15	19.90	4	75.48	-23.06	19.05	4.53	1.00
16	16.70	5	40.51				
17	13.10	3	23.61				
18	16.90	3	48.30				
19	17.60	7	55.99	-22.39	17.35	4.03	1.08
20	16.90	5	45.29	-24.32	17.98	4.22	0.84
21	19.50	5	73.30	-25.87	17.44	4.05	0.66
22	18.40	6	59.22				
23	8.10	1	5.65				
24	7.80	1	5.11				
25	8.00	1	5.89	-25.91	15.75	3.56	0.65
26	7.80	1	4.66				
27	8.00	1	5.16	-25.15	15.49	3.48	0.74
28	7.90	1	4.67	-25.53	15.40	3.45	0.70
29	8.10	1	5.11	-24.95	15.61	3.52	0.77
30	8.20	1	5.24	-26.33	16.13	3.67	0.60
31	7.60	1	5.32				
32	7.80	1	4.38				
33	7.80	1	4.46	-26.21	15.50	3.48	0.62
34	7.80	1	4.53	-25.66	15.87	3.59	0.68
35	7.60	1	4.53				
36	8.00	1	5.10	-26.52	15.63	3.52	0.58
37	8.60	1	6.62	-25.68	15.82	3.58	0.68
38	7.60	1	4.80				
39	8.10	1	5.68				
40	7.50	1	4.18	-26.23	15.64	3.53	0.61
41	17.00	5	49.56				
42	18.20	5	55.12	-25.77	18.35	4.32	0.67
43	15.50	3	38.24				
44	16.20	4	39.54	-24.22	16.29	3.72	0.86
45	16.60	3	41.51				
46	17.20	5	41.53	-25.11	18.63	4.40	0.75
47	17.00	5	47.78	-24.26	18.26	4.30	0.85
48	18.40	4	59.32				
49	17.40	5	50.44	-24.85	17.49	4.07	0.78
50	16.80	4	47.06				
51	19.00	5	53.73				
52	13.90	2	27.15	-23.01	16.65	3.82	1.00
53	15.40	3	34.85	-24.63	16.92	3.90	0.81
54	17.60	4	54.26				
55	16.10	4	36.12				
56	17.20	4	50.54	-23.51	17.57	4.09	0.94
57	7.70	1	4.38				
58	8.00	1	5.17				
59	7.80	1	4.88	-25.77	15.23	3.40	0.67
60	8.30	1	5.53	-25.66	15.04	3.35	0.68

OSISKO (OS)

FISH #	TOT L (cm)	age	WT (g)	d13C	d15N	TP litt	%ben
61	7.70	1	4.74				
62	7.00	1	3.57	-26.51	15.87	3.59	0.58
63	17.30	5	49.93	-24.38	17.33	4.02	0.84
64	12.40	3	21.42	-19.90	16.40	3.75	1.38
65	14.70	2	33.71	-24.54	16.91	3.90	0.82
66	14.30	3	30.59	-19.42	15.05	3.35	1.43
67	15.40	3	37.05				
68	14.70	3	39.73	-21.43	16.06	3.65	1.19
69	19.40	4	66.91				
70	18.10	4	61.92				
71	18.20	4	56.53	-22.08	16.96	3.91	1.11
72	7.50	1	3.57				
73	7.57	1	3.67				
74	8.00	1	4.33				
75	7.02	1	3.31				
76	6.58	1	2.50				
77	9.56	1	8.04				
78	7.01	1	2.76				
79	7.56	1	3.63				
80	7.57	1	3.62				
81	8.50	1	4.69				
82	8.01	1	4.35				
83	7.02	1	3.11				
84	8.03	1	4.76				
85	8.01	1	4.26				
86	7.58	1	3.50				
87	7.56	1	3.95				
88	7.57	1	3.39				
89	8.40	1	4.82				
90	7.40	1	3.01				
91	7.90	1	3.94				
os pool 1	7.97	1	5.24	-25.53	15.55	3.50	0.70
os pool 2	8.03	1	4.94	-25.83	15.75	3.56	0.66
os pool 3	7.65	1	4.36	-25.95	15.76	3.56	0.65
os pool 4	6.91	1	2.92	-25.49	15.12	3.37	0.70
os pool 5	7.58	1	3.59	-24.27	15.40	3.46	0.85
os pool 6	8.09	2	4.50	-24.75	14.91	3.31	0.79
os pool 7	9.03	1	6.37	-24.36	15.43	3.46	0.84
os pool 8	7.73	1	4.94	-25.51			0.70
os pool 9	7.65	1	4.59	-25.24			0.73
osis a	16.80	3	47.37	-23.09	14.40	3.16	0.99
osis c	16.20	5	42.04	-24.62	17.17	3.98	0.81
osis e	17.20	7	50.49	-23.48	18.27	4.30	0.94
osis g	16.60	6	45.02	-24.99	17.75	4.15	0.76
osis h	15.90	4	39.49	-23.77	17.02	3.93	0.91
osis i	16.10	5	43.49	-23.99	18.03	4.23	0.88
osis j	16.60	5	46.09	-25.64	17.49	4.07	0.68
osis l	18.70	5	39.63	-23.35	18.04	4.23	0.96
os yoy 1	1.51	0	0.05	-28.37	14.31	3.13	0.00
os yoy 2	1.51	0	0.05	-27.28	14.18	3.10	0.00
os yoy	1.51	0	0.05	-28.01	13.38	2.86	0.00

Appendix 1a. Perch length (TOTL), age, wet weight (WT), $\delta^{13}\text{C}$ (d13C) and $\delta^{15}\text{N}$ signature of white muscle tissue, trophic position (Tplitt – see methods for formula), and the proportion of benthivory in perch diet (%ben).

DUFAULT (DT)

FISH #	TOT L (cm)	AGE	WT (g)	d13C	d15 N	TP litt	%benth
1	15.10	4	36.33	-23.77	16.19	4.98	1.70
2	16.80	4	45.55	-23.47	17.18	5.27	1.76
3	16.90	5	47.96	-23.43	16.38	5.03	1.77
4	17.40	4	53.80	-20.68	16.76	5.14	2.36
5	10.50	2	9.82				
6	11.10	2	12.41				
7	11.70	3	12.33				
8	10.60	2	9.92				
9	10.70	2	9.54				
10	11.70	3	14.37				
11	10.70	2	10.83				
12	10.80	3	10.51				
13	12.00	3	14.52				
14	10.10	3	9.23				
15	10.90	3	11.92				
16	10.70	4	10.74				
17	10.30	2	9.89				
18	10.00	2	9.12				
19	11.10	3	10.78				
20	10.70	3	10.68				
21	10.10	4	9.35				
22	10.10	3	9.61				
23	10.50	3	10.56				
24	10.00	3	9.23				
25	9.90	2	9.75	-28.98	10.24	3.23	0.60
26	13.00	5	20.07	-29.87	10.80	3.39	0.41
27	11.90	4	13.35	-28.85	10.06	3.18	0.63
28	16.50	5	39.82	-29.05	10.43	3.28	0.58
29	16.40	7	41.71	-28.21	12.61	3.93	0.76
30	9.70	2	8.88				
31	11.50	3	13.86	-28.15	11.55	3.61	0.77
32	15.00	4	31.87	-29.41	10.57	3.32	0.51
33	15.30	5	31.15	-29.74	10.14	3.20	0.44
34	16.20	6	38.38	-29.21	10.41	3.28	0.55
35	12.00	4	17.98	-28.96	10.98	3.45	0.60
36	10.20	2	10.67				
37	10.50	2	11.85				
38	10.30	2	10.12				
39	9.90	2	10.08	-29.37	10.30	3.25	0.51
40	9.50	2	9.85	-29.47	10.65	3.35	0.49
41	15.70	7	34.63	-29.38	9.94	3.14	0.51
42	15.50	6	43.27	-28.32	12.37	3.85	0.74
43	12.10	4	13.58				
44	11.50	4	13.07	-28.83	10.77	3.38	0.63
45	11.00	5	11.06				
46	11.60	4	12.49	-27.08	10.95	3.44	1.00
47	14.00	5	24.01	-27.58	10.32	3.25	0.90
48	13.00	6	22.77				
49	14.20	5	27.84	-27.73	10.75	3.38	0.86
50	12.90	4	18.31				
51	13.80	4	24.80	-31.25	10.77	3.38	0.12
52	15.00	7	35.17	-29.57	11.99	3.74	0.47
53	12.70	4	27.02	-29.10	9.97	3.15	0.57
54	13.40	5	29.52	-28.19	10.35	3.26	0.76
55	5.30	1	1.23	-30.31	11.41	3.57	0.32
56	5.20	1	1.51	-29.89	10.88	3.42	0.40
57	6.10	1	1.89	-29.52	10.36	3.26	0.48
58	6.00	1	2.03	-30.18	11.03	3.46	0.34
59	7.10	1	3.56	-30.27	10.88	3.42	0.32

DUFAULT (DT)

FISH #	TOTL (cm)	AGE	WT (g)	d13C	d15 N	TP litt	%benth
60	7.00	1	3.47	-27.28	10.29	3.24	0.96
61	10.60	2					
62	13.90	4					
63	14.50	4					
64	10.60	2					
65	9.90	2					
66	13.80	4					
67	10.90	2					
68	6.90	1					
69	9.50	2		-28.93	10.57	3.32	0.61
70	11.20	3					
71	9.40	2					
72	9.50	2					
du pool 1	10.70	3	11.81	-28.57	10.90	3.42	0.68
du pool 2	10.50	3	11.46	-29.15	10.71	3.37	0.56
du pool 3	6.55	1	2.74	-29.32	10.64	3.35	0.53
du pool 4	6.10	1	1.23	-30.31	11.41	3.57	0.32
du pool 5	13.85	4		-27.08	10.71	3.37	1.00
du pool 6	10.70	2		-27.97	10.96	3.44	0.81
du pool 7	9.47	2		-28.93	10.57	3.32	0.61
du 1A	7.40	1	4.13	-29.13	9.64	3.05	0.57
du 1B	9.40	1	8.20	-28.85	9.51	3.01	0.62
du 1C	8.10	1	5.20	-29.06	9.87	3.12	0.58
du 1D	7.60	1	4.45	-29.17	10.27	3.24	0.56
du b	11.10	3	13.71	-27.59	11.00	3.45	0.89
du c	10.70	3	10.68	-28.80	11.40	3.57	0.63
du e	12.00	3	14.27	-27.65	10.93	3.43	0.88
du f	9.80	2	9.42	-29.08	10.58	3.33	0.58
du g	14.50	4	36.40	-29.87	12.28	3.83	0.41
du j	14.00	5	36.20	-29.62	10.42	3.28	0.46
du q	5.50	1	1.62	-30.00	11.67	3.65	0.38
du r	12.80	3	19.40	-27.27	11.03	3.46	0.96
du YOY	1.85	0	0.07	-31.68	8.23	2.64	0.03
du YOY	1.85	0	0.07	-28.26	13.46	4.18	0.03
du YOY	1.85	0	0.07	-27.61	13.87	4.29	0.03

Appendix 1b. Perch length (TOTL), age, wet weight (WT), $\delta^{13}\text{C}$ (d13C) and $\delta^{15}\text{N}$ signature of white muscle tissue, trophic position (Tplitt – see methods for formula), and the proportion of benthivory in perch diet (%ben).

OPASATICA (OP)

FISH #	TOTL (cm)	AGE	WT (g)	d13C	d15N	TP litt	%ben
1	14.10	4	30.28	-23.82	9.71	3.67	0.77
2	16.00	4	51.92	-22.90	9.52	3.62	0.85
3	19.50	5	92.53	-26.24	10.47	3.89	0.57
4	18.20	5	77.41	-25.38	10.20	3.81	0.64
5	19.40	4	104.52				
6	16.50	3	52.84				
7	23.70	5	162.56	-25.47	11.52	4.20	0.63
8	16.90	3	52.76	-30.74	9.19	3.52	0.19
9	19.20	5	96.35	-25.11	10.48	3.90	0.66
10	20.50	4	118.52	-24.86	10.31	3.85	0.69
11	18.60	3	75.32				
12	18.20	4	70.4				
13	15.00	3	35.63	-24.87	11.05	4.06	0.68
14	16.20	5	55.01				
15	23.00	6	161.93	-25.93	11.09	4.08	0.60
16	16.60	4	60.68				
17	17.10	3	66.04				
18	16.10	2	49.27				
19	14.70	2	35.81	-23.72	10.53	3.91	0.78
20	25.00	7	206.82	-25.11	10.61	3.94	0.66
21	18.60	5	82.59	-25.93	10.11	3.79	0.59
22	22.60	6	144.29	-25.71	11.72	4.26	0.61
23	18.60	8	81.48				
24	18.60	3	91.6				
25	21.30	4	137.4				
26	5.70	1	2.15	-27.38	8.84	3.42	0.47
27	6.10	1	2.58	-28.29	9.25	3.54	0.40
28	6.00	1	2				
29	6.20	1	2.33				
30	6.50	1	2.76	-26.33	8.47	3.31	0.56
31	5.30	1	1.55	-27.16	8.79	3.40	0.49
32	5.60	1	2.01				
33	5.90	1	2.19	-26.26	9.32	3.56	0.57
34	5.50	1	1.69				
35	5.40	1	1.77				
36	8.70	2	7.88	-25.79	8.93	3.44	0.61
37	8.90	2	7.25	-29.09	9.65	3.65	0.33
38	12.50	3	24.03				
39	12.90	3	24.71				
40	6.00	1	2.33				
41	16.30	3	55.98	-24.50	9.98	3.75	0.72
42	14.00	3	35.35	-21.14	9.11	3.49	1.00
43	6.60	1	2.66				
44	5.50	1	1.73				
45	6.20	1	2.46				
46	6.50	1	2.67				
47	6.40	1	2.72				
48	6.30	1	2.6				
49	19.90	5	102.97	-24.09	11.27	4.13	0.75
50	13.10	2	27.96	-24.27	10.37	3.87	0.74
51	14.20	3	32.1	-24.89	10.07	3.78	0.68
52	19.70	6	95.15	-25.81	12.12	4.38	0.60
53	20.40	4	107.87	-26.07	11.87	4.31	0.58
54	18.10	4	73.41				
55	25.00	9	196.06	-24.79	10.72	3.97	0.69
op pool 1	5.60		1.87	-26.71	9.06	3.48	0.53
op pool 2	6.30		2.67	-27.31	8.86	3.42	0.48
op pool 3	5.75	1	2.0275				
op pool 4	6.25	1	2.473333333				
op yoy				-26.04	10.73	3.97	0.00

Appendix 1c. Perch length (TOTL), age, wet weight (WT), $\delta^{13}\text{C}$ (d13C) and $\delta^{15}\text{N}$ signature of white muscle tissue, trophic position (Tplitt – see methods for formula), and the proportion of benthivory in perch diet (%ben).

DASSERAT (DA)

FISH #	TOT L (cm)	AGE	WT (g)	d15N	d13C	TP litt	% ben
1	13.50	2	30.05	8.00	-24.07	3.00	0.92
2	16.50	3	64.62	9.02	-24.19	3.30	0.90
3	25.20	5	215.97	9.70	-24.02	3.50	0.93
4	23.20	4	146.92	10.48	-23.46	3.73	1.00
5	19.70	3	105.30	9.52	-23.81	3.44	0.95
6	23.60	5	188.37	9.60	-23.93	3.47	0.94
7	12.70	3	26.21	9.00	-23.66	3.29	0.97
8	23.00	4	159.97	9.67	-23.72	3.49	0.97
9	24.50	5	182.44	9.35	-24.64	3.39	0.84
10	21.80	3	143.44	10.10	-24.82	3.61	0.82
11	22.50	4	163.87	10.58	-23.64	3.76	0.98
12	23.50	5	165.88				
13	10.40	1	14.35	8.82	-28.43	3.24	0.34
14	16.60	3	62.95				
15	13.50	3	35.21	8.93	-27.34	3.27	0.49
16	12.70	2	28.13	5.37	-32.00	2.22	
17	16.70	3	79.99	9.55	-25.82	3.45	0.69
18	14.60	2	45.59	9.18	-27.03	3.34	0.53
19	11.50	1	19.54	9.29	-26.49	3.38	0.60
20	13.00	2	29.41				
21	11.10	2	19.04	9.21	-26.70	3.35	0.57
22	15.00	2	45.36	8.87	-26.50	3.26	0.60
23	15.70	3	51.10	8.73	-25.14	3.21	0.78
24	20.20	5	126.04				
25	16.20	4	58.18				
26	18.60	4	89.26	9.34	-23.71	3.39	0.97
27	23.40	4	184.28	9.48	-24.89	3.43	0.81
28	22.20	4	144.89	9.94	-24.28	3.57	0.89
29	23.30	5	181.06	10.48	-24.57	3.73	0.85
30	17.70	3	76.59				
31	25.40	5	241.89	10.39	-24.58	3.70	0.85
32	23.30	7	169.65	9.69	-24.21	3.50	0.90
33	20.00	3	118.53				
34	20.20	3	128.84				
35	24.70	5	210.21	9.28	-25.72	3.37	0.70
36	26.80	8	263.48				
da yoy		0		8.09	-30.98	3.03	0.00
da yoy		0		8.18	-30.84	3.05	0.02
da yoy		0		8.01	-30.77	3.00	

Appendix 1d. Perch length (TOTL), age, wet weight (WT), $\delta^{13}\text{C}$ (d13C) and $\delta^{15}\text{N}$ signature of white muscle tissue, trophic position (Tplitt – see methods for formula), and the proportion of benthivory in perch diet (%ben).

VAUDRAY (VA)

FISH #	TOT L (cm)	AGE	WT (g)	d13C	d15N	TP litt	%benth
1	12.90	2	23.48	-24.49	7.92	2.97	0.97
2	11.90	2	18.44	-25.25	7.61	2.88	0.89
3	10.80	1	12.11	-25.27	8.01	3.00	0.89
4	10.20	2	12.17	-24.82	7.63	2.89	0.94
5	9.80	1	8.88	-25.61	7.62	2.89	0.85
6	8.40	1	4.48	-27.78	7.49	2.85	0.62
7	8.90	1	6.52	-27.13	7.86	2.96	0.69
8	9.50	1	7.38	-26.22	8.64	3.19	0.79
9	9.60	2	8.54	-26.68	7.76	2.93	0.74
10	9.70	1	10.30	-24.78	7.36	2.81	0.94
11	10.40	2	11.37	-25.43	7.81	2.94	0.87
12	12.50	2	19.96	-25.53	8.36	3.10	0.86
13	10.60	2	12.92	-25.50	6.48	2.55	0.86
14	10.40	2	11.83				
15	13.00	2	24.59	-25.34	7.84	2.95	0.88
16	16.00	3	46.32	-25.60	8.41	3.12	0.85
17	10.60	2	13.05	-25.62	7.61	2.88	0.85
18	17.20	4	56.57	-25.72	8.50	3.15	0.84
19	15.20	3	46.55	-24.23	8.10	3.03	1.00
20	14.30	3	32.41	-24.94	8.65	3.19	0.92
21	15.50	3	40.48	-25.84	8.86	3.25	0.83
22	15.10	4	37.55	-24.75	8.45	3.13	0.94
23	15.00	3	38.83	-25.22	8.59	3.17	0.89
24	12.90	3	23.56	-25.96			0.82
25	11.80	3	16.28	-26.04	8.70	3.20	0.81
26	13.00	3	24.15	-25.11	6.78	2.64	0.91
27	10.90	2	14.19				
28	12.30	3	20.42	-24.40	8.19	3.05	0.98
29	6.30	1					
30	6.40	1					
31	6.40	1					
32	6.50	1					
33	5.70	1					
34	6.40	1					
35	6.50	1					
36	6.40	1					
37	8.70	1					
38	8.90	1					
39	9.30	1					
40	8.80	1					
41	9.30	1					
va pool 1	6.00	1					
va pool 2	6.43	1					
va pool 3	9.10	1					
va pool 4	8.85	1					
va pool 5	10.40	2	12.61	-25.22	7.62	2.89	0.89
va pool 6	9.75	1	9.59	-25.19	7.49	2.85	0.90
va pool 7	9.25	3	7.53	-26.90	7.81	2.94	0.71

Appendix 1e. Perch length (TOTL), age, wet weight (WT), $\delta^{13}\text{C}$ (d13C) and $\delta^{15}\text{N}$ signature of white muscle tissue, trophic position (Tplitt – see methods for formula), and the proportion of benthivory in perch diet (%ben).

FISH #	TOT L (cm)	AGE	WT (g)	d13C	d15N	TP litt	%benth
1	9.30	1	9.38	-29.38			0.76
2	9.70	2	13.30	-28.04	8.06	3.15	1.06
3	8.90	1	7.08				
4	9.80	2	10.91				
5	7.80	1	5.92	-29.72	8.28	3.22	0.69
6	10.10	2	13.35				
7	10.20	2	11.77	-28.32	10.04	3.73	1.00
8	10.40	2	13.82				
9	10.60	2	14.38	-29.81	7.86	3.09	0.67
10	10.30	2	13.80	-29.24	8.59	3.31	0.79
11	9.50	2	9.63				
12	20.50	5	125.20	-29.18	9.04	3.44	0.81
13	14.70	2	47.18	-29.13	8.07	3.16	0.82
14	5.50	1	1.59	-30.72	9.43	3.56	0.46
15	5.10	1	1.50				
16	5.70	1	1.97	-31.02	8.78	3.37	0.40
17	6.20	1	2.65	-28.59	8.97	3.42	0.94
18	6.50	1	3.11				
19	6.70	1	3.09				
20	6.30	1	2.89				
21	7.00	1	3.51				
22	7.30	1	4.67	-30.88	8.22	3.20	0.43
23	12.60	3	25.72	-28.85	9.69	3.63	0.88
24	9.00	2	9.12				
25	12.50	3	26.05				
26	13.10	2	29.65				
27	18.90	7	84.15	-28.36	9.60	3.60	0.99
28	15.90	5	49.98	-29.30	9.57	3.60	0.78
29	25.40	8	207.70	-29.50	9.77	3.66	0.74
30	23.70	7	184.33	-29.50	9.38	3.54	0.74
31	23.50	7	160.58				
32	19.50	7	99.18				
33	20.80	5	106.51	-28.84	9.11	3.46	0.88
34	19.40	5	90.40				
35	22.10	8	160.25	-30.68	9.41	3.55	0.47
36	21.80	8	122.15				
37	17.30	4	70.84	-28.41			0.98
38	21.20	6	119.49	-29.76	9.46	3.56	0.68
39	24.10	7	196.56	-30.69	9.57	3.60	0.47
40	26.70	9	255.86	-29.10	9.71	3.64	0.83
bo pool 1	5.60	1	1.78	-30.87	9.11	3.46	0.43
bo pool 2	6.75	1	3.66	-29.74	8.60	3.31	0.68
bo pool 3	6.32	1	2.82				
bo pool 4	9.65	2	10.27				

BOUSQUET (BO)

Appendix 1f. Perch length (TOTL), age, wet weight (WT), $\delta^{13}\text{C}$ (d13C) and $\delta^{15}\text{N}$ signature of white muscle tissue, trophic position (Tplitt – see methods for formula), and the proportion of benthivory in perch diet (%ben).

APPENDIX 2
Stomach content data for perch.

frequency									
FISH #	totZOO	DIPTERA	TRICOP	HYDRACH	PELICYP	COLEOP	ODONATA	CRAYFISH	FISH
1	1	4	1	0	0	0	0	0	0
2	0	47	0	0	0	0	0	0	0
3	0	108	0	0	0	0	0	0	0
4	0	37	0	0	0	0	0	0	0
5	0	42	0	0	0	0	0	0	0
6	0	51	0	0	0	0	0	0	0
7	0	1	0	0	0	0	0	0	0
8	0	4	0	0	0	0	0	0	0
9	0	6	0	0	0	0	0	0	0
10	0	11	0	0	0	0	0	0	0
11	0	40	0	0	0	0	0	0	0
12	0	32	5	0	0	0	0	0	0
13	0	5	2	1	0	0	0	0	0
14	0	2	0	15	0	0	0	0	0
15	0	48	0	0	0	0	0	0	0
16	0	8	0	0	0	0	0	0	0
17	0	18	0	0	0	0	0	0	0
18	0	10	8	0	0	0	0	0	0
19	0	26	0	0	1	0	0	0	0
20	0	8	0	0	0	0	0	0	0
21	0	1	0	0	0	0	0	0	0
22	0	0	0	0	0	1	0	0	0
23	0	58	0	0	0	0	0	0	0
24	41	0	0	0	0	0	0	0	0
25	51	0	0	0	0	0	0	0	0
26	10	0	0	0	0	0	0	0	0
27	14	0	0	0	0	0	0	0	0
28	6	0	0	0	0	0	0	0	0
29	12	0	0	0	0	0	0	0	0
30	11	0	0	0	0	0	0	0	0
31	6	0	0	0	0	0	0	0	0
32	9	0	0	0	0	0	0	0	0
33	21	0	0	0	0	0	0	0	0
34	5	0	0	0	0	0	0	0	0
35	13	0	0	0	0	0	0	0	0
36	3	0	0	0	0	0	0	0	0
37	4	0	0	0	0	0	0	0	0
38	1	0	0	0	0	0	0	0	0
39	36	0	0	0	0	0	0	0	0
40	8	0	0	0	0	0	0	0	0
41	0	64	0	1	0	0	0	0	0
42	0	91	1	0	0	0	0	0	0
43	0	3	0	0	0	0	0	0	0
44	0	56	0	0	0	0	0	0	0
45	0	1	0	0	0	0	0	0	0
46	0	2	1	0	0	0	0	0	0
47	0	96	0	0	0	0	0	0	0
48	0	3	1	0	0	0	0	0	0
49	0	3	0	13	0	0	0	0	0
50	0	17	0	1	0	0	0	0	0
51	0	4	0	13	0	0	0	0	0
52	0	50	3	0	0	0	0	0	0
53	0	0	0	0	0	0	0	0	0
54	0	47	0	0	0	0	0	0	0
55	0	3	4	10	0	0	0	0	0

OSISKO (OS)

Appendix 2a. The frequency of various prey items in the stomach contents of perch.

FISH #	totZOO	DIPTERA	TRICOP	HYDRACH	PELICYP	COLEOP	ODONATA	CRAYFISH	FISH
56	0	126	0	0	0	0	0	0	0
57	0	1	0	0	0	0	0	0	0
58	2	0	0	0	0	0	0	0	0
59	1	0	0	0	0	0	0	0	0
60	20	0	0	0	0	0	0	0	0
61	0	0	0	0	0	0	0	0	0
62	6	0	0	0	0	0	0	0	0
63	0	5	0	0	0	0	0	0	0
64	0	0	0	0	0	0	0	0	0
65	0	1	0	0	0	0	0	0	0
66	0	0	0	0	0	0	0	0	0
67	0	7	5	1	0	0	0	0	0
68	0	2	1	0	1	0	2	0	0
69	0	0	2	0	0	0	0	0	0
70	0	1	0	0	0	0	0	0	0
71	0	0	0	0	0	0	0	0	0
os 72	1408	0	0	0	0	0	0	0	0
Os 73	277	0	0	0	0	0	0	0	0
Os 74	386	1	0	0	0	0	0	0	0
Os 75	285	0	0	0	0	0	0	0	0
Os 76	176	2	0	0	0	0	0	0	0
Os 77	1	0	0	0	0	0	0	0	0
Os 78	113	0	0	0	0	0	0	0	0
Os 79	65	6	0	0	0	0	0	0	0
Os 80	338	0	0	0	0	0	0	0	0
Os 83	499	0	0	0	0	0	0	0	0
Os 84	442	0	0	0	0	0	0	0	0
Os 86	42	0	0	0	0	0	0	0	0
Os 87	151	1	0	0	0	0	0	0	0
os 88	497	2	0	0	0	0	0	0	0
os 90	65	0	0	0	0	0	0	0	0

OSISKO (OS)

Appendix 2a. The frequency of various prey items in the stomach contents of perch.

fish #	frequency						
	totZOO	DIPTERA	TRICOP	HYDRACH	PELICYP	CRAYFISH	FISH
1	0	4	0	0	0	0	0
2	0	32	0	9	0	0	0
3	0	25	0	0	0	0	0
4	0	11	0	0	0	0	0
5	660	2	0	0	0	0	0
6	720	0	0	0	0	0	0
7	770	0	0	0	0	0	0
8	510	0	0	0	0	0	0
9	840	2	0	0	0	0	0
10	550	0	0	0	0	0	0
11	370	0	0	0	0	0	0
12	875	0	0	0	0	0	0
13	0	2	1	0	0	0	0
14	900	1	0	0	0	0	0
15	34	3	0	0	0	0	0
15	400	4	0	0	0	0	0
17	1150	0	0	0	0	0	0
18	1350	0	0	0	0	0	0
19	4	27	0	0	0	0	0
20	530	0	0	0	0	0	0
21	670	1	0	0	0	0	0
22	380	0	0	0	0	0	0
23	2000	0	0	0	0	0	0
24	160	0	0	0	0	0	0
25	2915	0	0	0	0	0	0
26	270	3	0	0	0	0	0
27	690	7	0	0	0	0	0
28	60	4	0	0	0	0	0
29	0	1	0	0	0	0	0
30	1000	0	0	0	0	0	0
31	31	2	0	0	0	0	0
32	320	15	0	0	0	0	0
33	64	11	0	0	0	0	0
34	200	6	0	0	0	0	0
35	300	5	1	0	0	0	0
36	320	0	0	0	0	0	0
37	720	1	0	0	0	0	0
38	1085	0	0	0	0	0	0
39	710	0	0	0	0	0	0
40	255	2	0	0	0	0	0
41	180	2	0	0	0	0	0
42	0	0	0	0	0	0	1
43	0	12	2	0	0	0	0
44	125	0	0	0	0	0	0
45	4	6	0	0	0	0	0

DUFAULT (DT)

Appendix 2b. The frequency of various prey items in the stomach contents of perch.

fish #	frequency						
	totZOO	DIPTERA	TRICOP	HYDRACH	PELICYP	CRAYFISH	FISH
46	0	16	7	0	0	0	0
47	0	10	4	11	0	0	0
48	0	15	0	0	0	0	0
49	0	9	2	0	0	0	0
50	0	30	0	0	0	0	0
51	50	0	0	0	0	0	0
52	0	6	24	0	2	0	0
53	0	82	0	0	0	0	0
54	0	5	1	0	0	0	0
55	20	0	0	0	0	0	0
56	12	1	0	0	0	0	0
57+58	7	0	0	0	0	0	0
59	182	0	0	0	0	0	0
60	322	2	0	0	0	0	0
Du 61	313	1	0	0	0	0	0
Du 64	740	0	0	0	0	0	0
Du 69	24258	0	0	0	0	0	0
Du 71	395	0	0	0	0	0	0
Du 72	467	3	0	0	0	0	0
Du A	5	9	0	0	0	0	0
Du C	1076	0	0	0	0	0	0
Du D	1296	0	0	0	0	0	0
Du G	0	14	0	0	0	0	1
Du J	0	68	0	0	0	0	0
Du K	0	0	0	0	0	0	0
Du L	1445	0	0	0	0	0	0

DEFAULT (DT)

Appendix 2b. The frequency of various prey items in the stomach contents of perch.

DASSERAT (DA)

FISH#	frequency											OTHER
	tot ZOOP	DIFE	TRICOP	PELICYP	COLEOP	ODON	EHEM	HEXAG	AMPHE	CRAYFISH	FISH	
1	0	0	0	0	0	3	1	0	1	0	0	1 noid
2	0	1	0	0	0	1	0	0	71	0	0	0
3	0	4	0	0	0	0	0	0	6	0	0	0
4	0	1	1	0	0	13	0	0	0	0	0	0
5	0	2	3	0	0	0	1	0	8	0	0	0
6	0	0	0	0	0	0	0	0	0	1	0	0
7	1	0	0	0	0	2	0	1	3	0	0	1 leach
8	2	0	1	0	0	1	1	0	11	0	0	0
9	0	1	2	0	0	0	0	0	5	0	0	undirt
10	0	0	0	0	0	3	0	0	20	0	0	0
11	0	0	0	1	2	2	0	0	2	0	0	0
12	0	1	1	0	0	3	0	0	5	0	0	0
13	0	0	1	0	0	2	0	0	0	0	3	1 leach
14	0	0	0	0	0	0	0	0	0	0	0	0
15	0	0	0	0	0	0	0	0	1	1	0	0
16	0	0	0	0	0	2	0	0	20	0	0	0
17	0	3	0	0	0	0	0	0	13	0	0	0
18	0	0	1	0	0	1	0	0	0	0	0	fishscale
19	0	0	0	0	0	0	0	0	0	1	0	0
20	0	0	0	0	0	0	0	0	0	1	0	0
21	0	0	1	0	0	3	0	0	0	0	0	0
22	0	0	0	1	0	0	0	0	1	0	0	2 leeches
23	0	0	0	0	0	3	0	0	6	0	0	0
24	0	1	0	0	0	1	0	0	0	0	0	1 leach
25	0	0	0	0	0	0	0	0	0	2	0	snail
26	0	0	0	0	0	2	0	0	1	0	0	leach
27	0	0	0	0	0	3	0	0	0	0	0	0
28	0	0	0	0	0	0	0	0	0	0	0	0
29	0	0	1	1	0	3	0	0	7	0	0	fishscales
30	0	0	0	0	0	0	0	0	0	0	0	0
31	0	0	1	0	0	3	0	0	3	0	0	0
32	0	0	9	0	0	0	0	0	2	0	0	0
33	0	0	0	0	0	1	0	0	0	0	2	0
34	0	0	0	0	0	1	0	0	0	0	0	0
35	0	0	0	0	0	0	0	0	0	1	0	snail
36	0	0	2	0	0	0	0	0	0	0	0	snail

Appendix 2c. The frequency of various prey items in the stomach contents of perch.

OPASATICA (OP)

FISH #	frequency								
	totZOO	DIPTERA	TRICOP	HYDRACH	ODON.	EPHEMER	AMPHIP.	CRAYFISH	FISH
1	0	0	0	0	8	3	0	0	0
2	0	0	0	0	6	7	2	0	0
3	0	0	0	0	0	0	0	0	1
4	0	0	0	0	0	0	0	0	5
5	0	0	0	0	0	0	0	0	0
6	0	0	12	0	0	1	0	0	0
7	0	0	0	0	0	0	0	0	1
8	0	0	0	0	0	1	0	0	1
9	0	0	0	0	0	0	0	0	4
10	0	0	0	0	0	0	0	0	2
11	0	1	0	0	0	0	0	0	0
12	0	0	0	0	2	0	0	0	0
13	0	1	0	24	0	1	0	0	0
14	0	0	0	0	0	0	0	0	0
15	0	0	0	0	0	0	0	0	0
16	0	0	0	0	0	0	0	0	0
17	0	0	0	0	1	0	0	0	0
18	0	1	8	0	0	0	4	0	0
19	0	0	0	0	2	1	0	0	1
20	0	0	0	0	0	0	0	0	2
21	0	0	0	0	0	0	0	0	0
22	0	0	0	0	0	0	0	0	0
23	0	0	0	0	0	0	0	0	0
24	0	0	0	0	4	7	0	0	0
25	0	0	0	0	0	0	0	0	1
26	120	1	0	0	0	0	0	0	0
27	1000	1	0	0	0	0	0	0	0
28	48	0	0	0	0	0	0	0	0
29	0	0	0	0	0	0	0	0	0
30	0	0	0	0	0	0	0	0	0
31	0	0	0	0	0	0	0	0	0
32	0	2	0	0	0	0	0	0	0
33	420	0	0	0	0	0	0	0	0
34	0	0	0	0	0	0	0	0	0
35	0	0	0	0	0	0	0	0	0
36	0	0	0	0	0	0	0	0	0
37	0	6	0	0	0	0	0	0	0
38	0	0	1	0	1	0	0	0	0
39	0	0	0	0	0	0	0	0	0
40	0	0	0	0	0	0	0	0	0
41	0	0	0	0	0	1	0	0	0
42	0	0	0	0	1	0	0	0	0
43	0	0	0	0	0	0	0	0	0
44	120	2	0	0	0	0	0	0	0
45	0	0	0	0	0	0	0	0	0
46	0	0	0	0	0	0	0	0	0
47	0	0	0	0	0	0	0	0	0
48	450	0	0	0	0	0	0	0	0
49	0	0	0	0	0	0	0	0	0
50	0	0	0	0	0	0	0	0	0
51	0	0	1	0	1	0	0	0	1
52	0	0	0	0	1	0	0	0	0
53	0	0	0	0	0	0	0	0	0
54	0	0	0	0	0	0	0	0	0
55	0	0	0	0	0	0	0	0	0

Appendix 2d. The frequency of various prey items in the stomach contents of perch

frequency							
FISH #	TOT ZOOP	DIPTERA	TRICOP	COLEOP	ODONATA	AMPHIPODS	HEXAGENIA
1	0	0	0	0	0	0	15
2	0	0	0	0	0	0	7
3	0	0	0	0	0	0	2
4	0	0	0	0	0	0	4
5	0	0	0	0	1	0	2
6	161	0	0	0	0	1	0
7	0	0	0	0	0	0	1
8	0	0	1	0	0	0	2
9	0	3	0	0	0	0	4
10	0	0	0	0	0	0	2
11	0	0	0	0	0	1	0
12	0	0	0	0	0	0	0
13	0	0	0	0	0	0	2
14	0	0	0	0	0	0	2
15	0	0	0	0	0	0	2
16	0	0	0	0	1	0	0
17	0	0	0	0	0	0	0
18	0	0	0	0	0	0	3
19	0	0	0	0	0	0	0
20	0	0	0	0	0	0	6
21	0	0	0	0	0	0	0
22	0	0	0	0	0	0	3
23	0	0	0	0	0	0	2
24	0	0	0	0	0	1	0
25	0	2	0	0	0	1	0
26	0	0	0	0	0	0	0
27	0	0	0	1	1	0	1
28	0	0	0	0	0	0	0
Va 29	1.66	1	0	0	0	0	0
Va 30	1.05	1	0	0	0	0	0
Va 31	2.14	3	0	0	0	0	0
Va 32	0	0	0	0	0	0	0
Va 34	4.76	2	0	0	0	0	0
Va 35	4.16	0	0	0	0	0	0
Va 36	3.78	1	0	0	0	0	0
Va 37	0	0	0	0	0	0	2
Va 38	5.11	3	0	0	0	0	0
Va 39	0	0	0	0	0	0	2

VAUDRAY (VA)

Appendix 2e. The frequency of various prey items in the stomach contents of perch

BOUSQUET (BO)

FISH #	frequency										
	toZOO	DIPTERA	TRICOP	HYDRACH	ODON.	EPHEM.	HEXAG.	AMPHIPODS	CRAYFISH	FISH	OTHER
1	0	1	0	2	0	1	2	0	0	0	0
2	0	4	0	0	0	0	2	7	0	0	3 hemiptera
3	3	2	1	0	0	0	0	0	0	0	0
4	0	0	0	0	0	0	4	0	0	0	0
5	300	3	0	0	0	2	0	1	0	0	0
6	0	0	0	0	0	0	3	0	0	0	0
7	0	0	0	0	0	0	2	0	0	0	0
8	0	4	0	0	0	0	1	1	0	0	0
9	0	3	0	0	0	0	2	70	0	0	2 hemiptera
10	0	0	0	0	0	0	0	4	0	0	no id
11	0	0	0	0	0	20	2	0	0	0	0
12	0	0	0	0	0	0	0	0	0	0	0
13	0	0	0	0	0	0	0	0	0	1	0
14	10	0	0	0	0	4	0	7	0	0	0
15	656	4	0	0	0	0	0	12	0	0	0
16	17	0	0	0	0	1	0	2	0	0	0
17	46	8	0	0	0	0	0	1	0	0	0
18	35	5	0	0	0	2	0	36	0	0	1 hemiptera
19	30	2	1	0	0	4	0	12	0	0	1 hemiptera
20	21	2	0	0	0	1	0	5	0	0	0
21	45	1	0	0	0	1	0	14	0	0	1 hemiptera
22	14	0	0	0	0	5	0	11	0	0	0
23	0	1	0	0	0	0	2	1	0	0	0
24	0	2	0	0	0	0	0	0	0	0	68 neurop., 2 hemip.
25	1	0	0	0	0	1	0	0	0	0	7 neurop., 2 hemip.
26	8	0	0	0	0	2	0	0	0	0	0
27	0	3	0	0	0	0	0	0	0	0	0
28	0	0	0	0	0	0	3	0	0	0	0
29	0	0	0	0	0	1	0	0	2	0	0
30	0	0	1	0	0	0	0	0	3	0	0
31	0	4	1	0	0	0	2	0	0	0	0
32	0	0	14	0	0	0	0	1	0	0	3 neurop
33	0	0	12	0	0	0	8	0	0	0	0
34	0	0	0	0	1	0	2	0	0	0	0
35	0	0	0	0	7	0	0	0	0	0	0
36	0	0	23	0	0	0	3	0	0	0	0
37	0	0	2	0	0	0	7	0	0	0	0
38	0	0	0	0	5	0	2	0	1	0	0
39	0	0	2	0	0	0	3	0	0	0	0

Appendix 2f. The frequency of various prey items in the stomach contents of perch

APPENDIX 3.
Mean concentration of metals in perch livers.

FISH #	Cu+	Cd+
OSISKO (OS)		
1	14.23	20.71
2	93.88	34.87
3	5.70	24.16
4	52.48	43.37
5	8.00	9.50
6	47.49	12.98
7	49.91	54.54
8	155.05	34.64
9	49.14	46.39
10	155.34	34.78
12	87.31	31.34
13	193.95	60.18
15	197.20	21.68
17	51.23	46.47
19	116.01	40.85
20	17.50	34.02
21	82.69	26.57
23	59.59	91.67
26	36.93	10.88
42	211.08	22.70
44	80.69	19.14
46	18.90	21.91
47	85.47	24.30
49	66.75	45.08
51	118.03	48.42
52	49.21	5.92
53	18.95	28.72
56	122.15	35.50
58	36.95	10.89
60	43.17	9.35
62	58.12	
63	27.85	17.45
64	39.07	11.62
65	74.29	1.36
66	296.92	14.37
68	224.54	19.60
70	28.85	19.67
os pool 1	39.96	3.24
os pool 2	44.90	11.02
os pool 3	24.06	7.09
os pool 4	13.58	1.76
os pool 5	9.42	1.66
os pool 6	3.00	0.88
os pool 7	6.16	1.81
os pool 8	29.50	16.40
os pool 9	19.40	46.10

FISH #	Cu+	Cd+
OSISKO (OS)		
osis a	35.50	25.26
osis c	43.30	34.32
osis e	70.59	46.25
osis g	39.08	28.97
osis h	186.39	31.14
osis i	22.68	22.75
osis j	19.93	27.86
osis l	98.46	22.35
os yoy 2	2.66	0.49

DUFAULT (DT)		
1	46.78	24.22
2	34.76	35.35
3	32.85	42.14
4	261.79	32.17
9	13.90	8.39
16	27.35	8.06
26	139.60	34.97
27	37.65	12.32
28	99.04	47.48
29	50.88	70.60
32	85.26	41.13
33	277.41	89.48
34	214.94	112.20
35	233.37	48.52
39	12.86	22.32
41	180.98	67.43
42	35.91	28.17
45	106.02	20.62
46	242.57	47.63
47	204.17	66.53
49	126.87	80.09
51	7.63	27.62
52	42.71	72.43
53	60.34	23.30
54	118.06	63.20
63	53.99	6.52
65	35.50	10.46
70	31.32	9.23
du pool 1	77.82	24.97
du pool 2	162.47	41.82

FISH #	Cu+	Cd+
DUFAULT (DT)		
du pool 3	102.30	6.63
du pool 5	63.55	5.57
du pool 6	13.07	4.85
du pool 7	13.52	3.98
du 1A	11.40	11.00
du 1B	22.70	37.60
du 1C	6.20	4.20
du 1D	18.50	20.10
du b	109.48	13.88
du c	50.68	19.03
du e	200.33	12.34
du f	48.09	30.03
du g	27.65	72.87
du j	33.88	25.39
du r	136.48	44.94
du YOY	11.80	2.50

OPASATICA (OP)

1	5.04	1.48
2	8.17	3.65
3	11.26	6.55
4	7.05	1.27
5	9.94	4.20
6	11.69	5.16
7	13.43	4.85
8	12.00	4.50
9	13.26	4.07
10	9.33	1.81
11	9.18	3.83
12	7.16	1.79
13	10.20	1.26
14	6.44	1.69
15	9.86	1.60
18	15.16	
20	6.66	1.06
21	5.92	3.94
22	9.77	5.68
23	2.82	
24	7.79	1.25
36	30.47	8.97
37	25.85	7.61
39	10.05	6.41
41	9.76	1.82
42	1.42	
49	10.00	2.28
50	5.44	1.60

FISH #	Cu+	Cd+
OPASATICA (OP)		
51	10.11	2.98
52	15.77	8.88
53	10.66	0.56
55	24.65	7.40
op pool 2	24.39	
op pool 3	6.50	1.70
op pool 4	8.70	2.10

DASSERAT (DA)

1	4.67	1.38
2	15.53	3.41
4	17.00	4.51
5	36.56	4.69
6	12.31	3.44
7	4.40	1.90
8	17.59	8.97
9	24.79	8.10
11	26.14	5.10
12	14.94	5.26
13	8.42	2.48
15	2.55	0.99
16	10.44	1.22
17	9.85	1.77
18	0.28	0.92
19	14.17	1.76
21	7.15	2.11
22	14.72	5.71
23	12.61	5.07
26	15.52	3.28
27	12.12	2.94
28	10.38	3.03
29	25.92	3.77
32	14.02	5.43
34	0.28	0.12
35	12.90	3.23
da YOY	0.28	

VAUDRAY (VA)

1	9.30	34.04
2	17.95	13.54
3	10.88	41.69
6	27.49	8.10
8	15.36	4.52
11	13.30	9.28
12	23.55	12.54
13	7.71	4.40
15	18.29	18.41

FISH #	Cu+	Cd+
VAUDRAY (VA)		
16	22.85	20.65
18	16.69	17.30
19	20.50	15.60
20	14.61	10.78
21	19.66	11.76
22	15.16	14.99
23	15.81	16.66
24	17.80	24.01
25	12.08	27.87
26	13.86	19.88
28	9.03	20.79
va pool 2	13.30	3.92
va pool 3	9.34	5.42
va pool 4	8.31	2.45
va pool 5	23.02	30.89
va pool 6	19.77	23.58
va pool 7	20.25	10.60
BOUSQUET (BO)		
1	34.16	10.06
2	15.88	4.68
3	36.21	10.67
7	13.04	3.84
9	13.01	3.83
10	11.30	3.33
12	19.73	16.40
13	12.63	5.85
23	18.07	9.88
24	3.50	2.60
27	9.86	16.76
28	17.04	15.60
29	31.18	22.14
30	100.06	21.37
33	15.01	17.57
35	10.91	17.08
37	3.99	3.95
38	25.37	17.08
39	19.24	19.40
40	19.94	17.91
bo pool 1	79.87	
bo pool 2	22.44	6.61
bo pool 3	10.80	5.20
bo pool 4	10.50	15.70

Appendix 3. The mean concentration of Cd⁺ and Cu⁺ in perch livers. Values are in µg/g dry weight.

APPENDIX 4:
Invertebrate abundance/m²

APPENDIX 4:
Invertebrate abundance/m²

OSISKO

ORDER/FAMILY	1A	1B	3A	4A	5A	5B	7A	9B	10A
Amphipoda	0.0	0.0	0.0	0.0	0.0	3.1	0.0	3.1	6.2
Talitridae	0.0	0.0	0.0	0.0	0.0	3.1	0.0	0.0	0.0
Coleoptera	0.0	0.0	0.0	0.0	0.0	18.6	3.1	12.4	6.2
Dytiscidae	0.0	0.0	0.0	0.0	0.0	6.2	0.0	0.0	3.1
Elmidae	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Halplidae	0.0	0.0	0.0	0.0	0.0	12.4	0.0	12.4	3.1
Diptera	711.1	1111.1	2355.6	622.2	40.4	195.7	59.0	65.2	406.8
Ceratopogonidae	0.0	0.0	0.0	0.0	0.0	3.1	6.2	0.0	0.0
Chaoborus	0.0	44.4	44.4	44.4	0.0	0.0	0.0	0.0	0.0
Chironomidae	711.1	1066.7	2311.1	577.8	80.7	192.5	52.8	65.2	406.8
Ephemeroptera	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Caenidae	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Ephemeridae	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Ephemerillidae	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Hexagenia	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Gastropoda	0.0	0.0	0.0	0.0	6.2	3.1	0.0	0.0	0.0
Hydrobiidae	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Lymnaeidae	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Planorbidae	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Valvatidae	0.0	0.0	0.0	0.0	0.0	9.3	0.0	0.0	0.0
Hempitera	0.0	0.0	0.0	0.0	0.0	6.2	0.0	0.0	0.0
Corixidae	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Hydrachnidia	0.0	0.0	44.4	0.0	21.7	28.0	0.0	0.0	55.9
Arrenuridae	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Hydrodromidae	0.0	0.0	0.0	0.0	0.0	9.3	0.0	0.0	0.0
Limnesiidae	0.0	0.0	0.0	44.4	0.0	0.0	0.0	0.0	12.4
Lymnocharidae	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Oxidae	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	9.3
Lepidoptera	0.0	0.0	0.0	0.0	15.5	3.1	3.1	0.0	0.0
Pyralidae	0.0	0.0	0.0	0.0	15.5	3.1	3.1	0.0	0.0
Odonata	0.0	0.0	0.0	0.0	6.2	0.0	3.1	12.4	0.0
Aeshnidae	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Coenagrionidae	0.0	0.0	0.0	0.0	6.2	0.0	3.1	12.4	0.0
Corduliidae	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Gomphidae	44.4	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Pelecypoda	0.0	0.0	88.9	0.0	71.4	59.0	0.0	74.5	2447.2
Sphaeridae	0.0	0.0	88.9	0.0	0.0	59.0	0.0	0.0	2447.2
Trichoptera	0.0	0.0	0.0	0.0	0.0	15.5	15.5	0.0	28.0
Leptoceridae	0.0	0.0	0.0	0.0	0.0	3.1	15.5	0.0	28.0
Molannidae	0.0	0.0	0.0	0.0	0.0	12.4	0.0	0.0	0.0
Polycentropodidae	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Sialis	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0

DFAULT												
ORDER/FAMILY	1B	5A	5B	7A	8A	8B	9B	10A	10B	11B	12A	12B
Amphipoda	6.2	6.2	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Talitridae	6.2	6.2	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Coleoptera	6.2	6.2	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Elmidae	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Haliplidae	6.2	6.2	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Diptera	397.5	142.9	260.9	65.2	692.5	195.7	311.1	133.3	222.2	488.9	177.8	44.4
Ceratopogonidae	0.0	0.0	0.0	0.0	3.1	195.7	0.0	0.0	0.0	0.0	0.0	0.0
Chaoborus	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Chironomidae	397.5	142.9	260.9	62.1	689.4	0.0	311.1	133.3	222.2	0.0	177.8	44.4
Athericidae	0.0	0.0	0.0	3.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Ephemeroptera	260.9	80.7	130.4	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Caenidae	260.9	80.7	130.4	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Ephemeridae	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Ephemerillidae	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Gastropoda	0.0	6.2	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Hydrobiidae	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Lymnaeidae	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Planorbidae	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Valvatidae	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Hemiptera	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Corixidae	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Hydrachnidia	6.2	24.8	49.7	3.1	3.1	9.3	0.0	0.0	0.0	0.0	44.4	0.0
Hydrachnidae	0.0	0.0	0.0	0.0	0.0	9.3	0.0	0.0	0.0	0.0	0.0	0.0
Arrenuridae	6.2	24.8	49.7	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Hydrodromidae	0.0	0.0	0.0	0.0	3.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Limnesiidae	0.0	0.0	0.0	3.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Lymnocharidae	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Oxidae	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Lepidoptera	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Pyalidae	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Odonata	0.0	6.2	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Aeshinidae	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Coenagrionidae	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Corduliidae	0.0	6.2	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Gomphidae	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Pelecypoda	335.4	111.8	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Sphaeridae	335.4	111.8	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Trichoptera	18.6	31.1	68.3	31.1	80.7	15.5	0.0	0.0	0.0	0.0	0.0	0.0
Leptoceridae	6.2	24.8	68.3	31.1	80.7	15.5	0.0	0.0	0.0	0.0	0.0	0.0
Molannidae	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Phryganeidae	12.4	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Polycentropodidae	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Psychomiidae	0.0	6.2	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0

OPASATICA

ORDER/FAMILY	2A	2B	3B	4B	5A	5B	8A	9A	10A	11B
Amphipoda	0.0	3.1	0.0	3.1	3.1	0.0	13.0	177.8	0.0	0.0
Talitridae	0.0	3.1	0.0	0.0	3.1	0.0	13.0	177.8	0.0	0.0
Coleoptera	3.1	6.2	3.1	0.0	0.0	0.0	40.4	0.0	0.0	0.0
Elmidae	3.1	0.0	3.1	0.0	0.0	0.0	40.4	0.0	0.0	0.0
Haliplidae	0.0	6.2	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Diptera	385.1	208.1	220.5	80.7	357.1	465.8	0.0	1200.0	444.4	888.9
Ceratopogonidae	0.0	3.1	3.1	0.0	9.3	77.6	0.0	133.3	88.9	0.0
Chironomidae	385.1	204.9	217.4	77.6	347.8	332.3	239.1	1066.7	355.6	888.9
Tabanidae	0.0	0.0	0.0	3.1	0.0	0.0	18.6	0.0	0.0	0.0
Athericidae	0.0	0.0	0.0	0.0	0.0	0.0	220.5	0.0	0.0	0.0
Ephemeroptera	59.0	21.7	0.0	43.5	804.3	49.7	0.0	577.8	355.6	0.0
Caenidae	59.0	21.7	0.0	0.0	801.2	43.5	0.0	488.9	311.1	0.0
Ephemeridae	0.0	0.0	0.0	0.0	3.1	3.1	167.7	44.4	44.4	0.0
Ephemerillidae	0.0	0.0	0.0	0.0	0.0	3.1	167.7	44.4	0.0	0.0
Gastropoda	43.5	124.2	68.3	161.5	559.0	506.2	0.0	177.8	44.4	0.0
Hydrobiidae	43.5	102.5	37.3	146.0	468.9	419.3	0.0	0.0	0.0	0.0
Lymnaeidae	0.0	0.0	0.0	0.0	3.1	0.0	583.9	0.0	0.0	0.0
Planorbidae	0.0	15.5	0.0	12.4	21.7	74.5	546.6	133.3	0.0	0.0
Valvatidae	0.0	6.2	31.1	0.0	65.2	12.4	0.0	44.4	44.4	0.0
Hemiptera	0.0	0.0	0.0	9.3	24.8	18.6	28.0	44.4	0.0	0.0
Corixidae	0.0	0.0		0.0	24.8	18.6	9.3	44.4	0.0	0.0
Hydrachnidia	3.1	18.6	6.2	0.0	136.6	96.3	18.6	133.3	0.0	0.0
Hydrachnidae	0.0	0.0		0.0	0.0	0.0	18.6	0.0	0.0	0.0
Arrenuridae	3.1	18.6	3.1	0.0	68.3	6.2	15.5	44.4	0.0	0.0
Hydrodromidae	0.0	0.0		0.0	31.1	24.8	0.0	0.0	0.0	0.0
Limnesiidae	0.0	0.0	0.0	0.0	0.0	31.1	3.1	88.9	0.0	0.0
Lymnocharidae	0.0	0.0	3.1	0.0	18.6	15.5	3.1	0.0	0.0	0.0
Oxidae	0.0	0.0	0.0	0.0	18.6	18.6	0.0	0.0	0.0	0.0
Lepidoptera	0.0	0.0	0.0	0.0	0.0	3.1	6.2	0.0	0.0	44.4
Pyalidae	0.0	0.0	0.0	0.0	0.0	3.1	3.1	0.0	0.0	44.4
Odonata	3.1	6.2	0.0	12.4	31.1	21.7	0.0	0.0	0.0	0.0
Aeshinidae	0.0	0.0	0.0	0.0	0.0	3.1	0.0	0.0	0.0	0.0
Coenagrionidae	3.1	0.0	0.0	3.1	24.8	24.8	12.4	0.0	0.0	0.0
Corduliidae	0.0	0.0	0.0	0.0	3.1	3.1	0.0	0.0	0.0	0.0
Gomphidae	0.0	9.3	0.0	9.3	6.2	0.0	6.2	0.0	0.0	0.0
Pelecypoda	83.9	59.0	21.7	93.2	71.4	394.4	0.0	0.0	88.9	88.9
Sphaeridae	0.0	0.0	0.0	0.0	0.0	0.0	6.2	0.0	88.9	88.9
Trichoptera	24.8	31.1	0.0	0.0	12.4	15.5	242.2	177.8	0.0	0.0
Leptoceridae	18.6	18.6	0.0	0.0	9.3	15.5	242.2	177.8	0.0	0.0
Molannidae	3.1	0.0	0.0	0.0	0.0	0.0	49.7	0.0	0.0	0.0
Phryganeidae	0.0	0.0	0.0	0.0	0.0	0.0	49.7	0.0	0.0	0.0
Polycentropodidae	0.0	0.0	0.0	0.0	3.1	0.0	0.0	0.0	0.0	0.0
Psychomyiidae	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0

OPASATICA

ORDER/F/ 1A	3A	6A	7A	7B	8A	8B	
Amphipoda	0.0	0.0	0.0	12.4	3.1	40.4	0.0
Talitridae							
Coleoptera	0.0	9.3	0.0	0.0	0.0	0.0	0.0
Elmidae							
Haliplidae							
Diptera	74.5	428.6	68.3	229.8	12.4	239.1	434.8
Ceratopogonidae							
Chironomidae							
Tabanidae							
Athericidae							
Ephemeroptera	34.2	31.1	15.5	3.1	0.0	167.7	90.1
Caenidae							
Ephemeridae							
Ephemerillidae							
Gastropoda	21.7	347.8	326.1	658.4	1062.1	583.9	478.3
Hydrobiidae							
Lymnaeidae							
Planorbidae							
Valvatidae							
Hemiptera	0.0	0.0	0.0	0.0	0.0	18.6	18.6
Corixidae							
Hydrachnia	3.1	15.5	9.3	3.1	28.0	15.5	21.7
Hydrachnidae							
Arrenuridae							
Hydrodromidae							
Limnesiidae							
Lymnocharidae							
Oxidae							
Lepidoptera	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Pyralidae	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Odonata	0.0	6.2	3.1	0.0	15.5	12.4	0.0
Aeshnidae							
Coenagrionidae							
Corduliidae							
Gomphidae							
Pelecypoda							
Sphaeriidae	46.6	59.0	31.1	21.7	37798.1	242.2	18.6
Trichoptera							
Leptocerida	18.6	133.5	3.1	9.3	0.0	49.7	31.1
Molannidae							
Phryganeidae							
Polycentropodidae							
Psychomyiidae							

DASSERAT

ORDER/FAMIL'	1A	1B	2A	2B	4B	5Atot	6A	7A	8A	10A/B
Amphipoda	158.4	43.5	201.9	177.0	21.7	52.8	524.8	46.6	111.8	711.1
Talitridae										
Coleoptera	3.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Elmidae										
Haliplidae										
Diptera	313.7	28.0	518.6	1944.1	267.1	425.5	583.9	198.8	1944.1	6044.4
Ceratopogonidae										
Chironomidae										
Tabanidae										
Athericidae										
Ephemeroptera	3.1	3.1	0.0	6.2	0.0	6.2	6.2	3.1	31.1	844.4
Caenidae										
Ephemeridae										
Ephemerillidae										
Gastropoda	267.1	152.2	528.0	599.4	3.1	49.7	524.8	180.1	332.3	1111.1
Hydrobiidae										
Lymnaeidae										
Planorbidae										
Valvatidae										
Hemiptera	0.0	0.0	0.0	6.2	0.0	0.0	0.0	0.0	0.0	0.0
Corixidae										
Hydrachnidia	62.1	49.7	177.0	121.1	21.7	9.3	40.4	62.1	127.3	133.3
Hydrachnidae										
Arrenuridae										
Hydrodromidae										
Limnesiidae										
Lymnocharidae										
Oxidae										
Lepidoptera	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Pyralidae										
Odonata	9.3	0.0	0.0	0.0	0.0	0.0	6.2	6.2	3.1	0.0
Aeshinidae										
Coenagrionidae										
Corduliidae										
Gomphidae										
Pelecypoda	732.9	62.1	1024.8	975.2	74.5	18.6	142.9	301.2	909.9	7866.7
Sphaeriidae										
Trichoptera	254.7	201.9	118.0	167.7	77.6	37.3	152.2	59.0	220.5	400.0
Leptoceridae										
Molannidae										
Phryganeidae										
Polycentropodidae										
Psychomiidae										

DASSERAT

ORDER/FAMILY	11A/B	12A	12B
Amphipoda	2533.3	0.0	44.4
Talitridae			
Coleoptera	0.0	0.0	0.0
Elmidae			
Haliplidae			
Diptera	1777.8	2044.4	1955.6
Ceratopogonidae			
Chironomidae			
Tabanidae			
Athericidae			
Ephemeroptera	88.9	177.8	133.3
Caenidae			
Ephemeridae			
Ephemerillidae			
Gastropoda	2311.1	0.0	177.8
Hydrobiidae			
Lymnaeidae			
Planorbidae			
Valvatidae			
Hemiptera	0.0	0.0	0.0
Corixidae			
Hydrachnidia	266.7	44.4	88.9
Hydrachnidae			
Arrenuridae			
Hydrodromidae			
Limnesiidae			
Lymnocharidae			
Oxidae			
Lepidoptera	0.0	0.0	0.0
Pyalidae			
Odonata	88.9	0.0	0.0
Aeshinidae			
Coenagrionidae			
Corduliidae			
Gomphidae			
Pelecypoda	1955.6	533.3	88.9
Sphaeridae			
Trichoptera	88.9	44.4	88.9
Leptoceridae			
Molannidae			
Phryganeidae			
Polycentropodidae			
Psychomiidae			

VAUDRAY

ORDER/FAMILY	1A	2A	2B	3A	4B	5B	6A	8B	1B	11B	12A
Amphipoda	6.2	245.3	524.8	18.6	419.3	0.0	264.0	3.2	0.0	0.0	0.0
Talitridae											
Coleoptera	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Dytiscidae											
Elmidae											
Haliplidae											
Diptera	153.2	49.7	19.3	59.6	226.8	56.0	872.7	295.3	44.4	888.9	177.8
Ceratopogonidae											
Chaoborus											
Chironomidae											
Ephemeroptera	158.4	118.1	149.7	6.2	6.2	3.2	13.4	0.0	88.9	0.0	0.0
Caenidae											
Ephemeridae											
Ephemerillidae											
Hexagenia											
Gastropoda	121.1	587.0	397.5	9.3	59.6	15.5	53.2	21.7	0.0	711.1	222.2
Hydrobiidae											
Lymnaeidae											
Planorbidae											
Valvatidae											
Hemiptera	0.0	0.0	0.0	0.0	0.0	0.0	9.3	0.0	0.0	0.0	0.0
Corixidae											
Hydrachnidia	37.3	4.4	62.1	6.2	43.5	0.0	49.7	0.0	0.0	44.4	0.0
Arrenuridae											
Hydrodromidae											
Limnesiidae											
Lymnocharidae											
Oxidae											
Lepidoptera	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Pyralidae											
Odonata	6.2	9.3	18.6	0.0	0.0	9.3	34.2	0.0	0.0	88.9	0.0
Aeshinidae											
Coenagrionidae											
Corduliidae											
Gomphidae											
Pelecypoda	56.0	18.7	127.3	31.6	214.3	24.8	52.8	24.8	0.0	577.8	88.9
Sphaeriidae											
Trichoptera	28.0	59.6	136.6	12.4	46.6	6.2	152.2	3.2	0.0	133.3	0.0
Leptoceridae											
Molannidae											
Polycentropodidae											