# Metals and Metallothionein in Fishes and Metals in Sediments from Lakes Impacted by Uranium Mining and Milling in Northern Saskatchewan

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## ABSTRACT

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This study evaluated the impacts of metals on large-bodied fishes and sediments from waters receiving effluents and mine dewatering discharges released at the Key Lake uranium mine site in north-central Saskatchewan. Sediments and fishes from lakes receiving either treated mill effluent or mine dewatering discharges were analyzed for As, Cd, Co, Cu, Fe, Hg, Mn, Mo, Ni, Pb, Se and Zn. Comparisons were made to fishes from reference lakes in the vicinity of the McArthur River and Cigar Lake mine sites. The framework for the study was the use of Guidance Questions proposed by the Aquatic Effects Technology Evaluation (AETE) program for consideration in developing an Environmental Effects Monitoring (EEM) program for Canadian metal mines as prescribed under the Metal Mining Effluent Regulations (MMER) of the ***Fisheries Act***. The magnitude and extent of metal contamination in lakes receiving effluents at the Key Lake mine site were evaluated through analyses of sections of sediment cores. Enrichment Factors (EFs), the ratios of metal concentrations in the top 1-cm section of core to those concentrations in the deeper (pre-operational) 20 cm section, demonstrated the highest degree of contamination for As, Mo and Se in lakes receiving treated mill effluent, and for Co and Ni in a lake receiving mine dewatering discharges. The bioavailability of all metals to white suckers, northern pike and lake whitefish was determined by analyses of their livers and kidneys. Responses to Cd, Cu, Hg, and Zn were evaluated by analyzing for the metal-binding protein, metallothionein (MT). Comparison of these analyses to those conducted on livers and kidneys of fishes from reference lakes demonstrated effluent-dependent and fish species-dependent bio-availabilities. In Fox Lake, which receives treated mill effluent, white suckers had the highest (6 to 38 times reference levels) hepatic and renal Mo and Se concentrations. In these white suckers, hepatic As (>15 times) and renal Ni (> 5 times) were highest; whereas hepatic Cu, Zn, Cd, Hg and MT concentrations were the lowest when compared to concentrations in white suckers from reference lakes. In Little McDonald Lake, receiving mine dewatering discharges, hepatic and renal Mo, Ni, Co and Cd in northern pike were the highest (1.5 to 43 times higher than concentrations in northern pike from reference lakes). No differences were observed in renal MT concentrations in white suckers, and in renal and hepatic MT concentrations in northern pike. The main body of the report concludes by presenting a summary of the results according to the Guidance Questions and recommendations for additional research.

In the Appendix of this report, results are presented for concentrations of Cr, U, V in sections of sediment cores from the impacted lakes at the Key Lake mining site, and of Al, As, Ba, Cd, Co, Cr, Cu, Fe, Pb, Mn, Mo, Se, U, V and Zn from sites in Wollaston Lake that are influenced by discharges from the Rabbit

Lake mine site in north-eastern Saskatchewan. Sediment EFs were greatest for Cr and V in Wolf Lake, while U enrichment was greatest in surficial sediments in Little McDonald Lake. The bio-availability to white suckers and northern pike of Cr, U and V could not be determined due to the lack of sufficient tissue. At affected sites in Hidden Bay of Wollaston Lake, EFs were highest for As, Cd, Cu, Mn, Mo, Se, and U. Fishes were not collected from Wollaston Lake. Additional research using the AETE Guidance Questions is also recommended for the Rabbit Lake mine site.

Key-words: uranium mining; metallothionein; metals; fish; sediments.

## RÉSUMÉ

Klaverkamp, J.F., C.L. Baron, B.W. Fallis, C.R. Ranson, K.G. Wautier, and P. Vanriel. 2002. Metals and metallothionein in fishes and metals in sediments from lakes impacted by uranium mining and milling in northern Saskatchewan. Can. Tech. Rep. Fish. Aquat. Sci. 2420 : v + 72 p.

Dans la présente étude, nous avons évalué l'impact de métaux sur les grands poissons et les sédiments dans les lacs qui reçoivent les effluents et les eaux d'égouttage de la mine d'uranium du lac Key, située dans le centre-nord de la Saskatchewan. Nous avons dosé les éléments As, Cd, Co, Cu, Fe, Hg, Mn, Mo, Ni, Pb, Se et Zn dans les sédiments et les poissons des lacs recevant des effluents traités de l'usine de concentration du minerai (ci-après « effluents traités ») ou des eaux d'égouttage. Nous avons comparé les résultats avec ceux obtenus pour les poissons des lacs de référence, situés près de la mine de la rivière McArthur et de celle du lac Cigar. L'étude avait pour cadre les questions proposées dans le Programme d'évaluation des techniques de mesure d'impacts en milieu aquatique (ETIMA) afin d'élaborer le Programme d'études de suivi des effets sur l'environnement (ESEE) pour les mines de métaux canadiennes, conformément au *Règlement sur les effluents des mines de métaux* (*REMM*) pris en vertu de la ***Loi sur les pêches***. Nous avons évalué le niveau et l'étendue de la contamination par les métaux des lacs recevant les effluents de la mine du lac Key en analysant des carottes de sédiments. Les facteurs d'enrichissement (FE), soit les rapports entre les concentrations des métaux dans la couche supérieure de la carotte (le premier cm) et les concentrations à une profondeur de 20 cm (antérieure à l'exploitation minière), montrent que les plus fortes contaminations au As, au Mo et au Se se sont produites dans les lacs recevant des effluents traités, et que le Co et le Ni contaminent davantage un lac recevant les eaux d'égouttage. Nous avons déterminé la biodisponibilité de tous les métaux pour le meunier noir, le grand brochet et le grand corégone en analysant leur foie et leurs reins. Les réactions des poissons au Cd, au Cu, au Hg et au Zn ont été évaluées en déterminant la teneur en métallothionéine (MT), une protéine liant les métaux, de ces organes. La comparaison des résultats obtenus à ceux de l'analyse du foie et des reins des poissons des lacs de référence montre que la biodisponibilité des divers métaux dépend des effluents et des espèces de poissons. Dans le lac Fox, qui reçoit des effluents traités, le meunier noir a les concentrations de Mo et de Se les plus élevées dans le foie et les reins (de 6 à 38 fois les valeurs pour les lacs de référence). Cette espèce a également les teneurs les plus élevées en As dans les tissus hépatiques (>15 fois) et en Ni dans les tissus rénaux (> 5 fois), tandis que les concentrations de Cu, Zn, Cd, Hg et MT sont plus basses que chez le meunier noir des lacs de référence. Dans le Petit lac McDonald, recevant des eaux d'égouttage, le grand brochet présente les concentrations de Mo, Ni, Co et Cd hépatiques et rénales les plus élevées, soit de 1,5 à 43 fois celles des grands brochets des lacs de référence. Les concentrations de MT dans les reins du meunier noir et dans le foie et les reins du grand brochet ne sont pas différentes entre les lacs. Nous concluons la principale partie de notre rapport en présentant un sommaire des résultats en fonction des questions posées dans l'ETIMA et en proposant des voies à suivre en matière de recherche.

Dans l'annexe du présent rapport, nous présentons les concentrations de Cr, d'U et de V dans les carottes de sédiments des lacs recevant les effluents traités et les eaux usées de la mine du lac Key. Nous présentons également les concentrations d'Al, As, Ba, Cd, Co, Cr, Cu, Fe, Pb, Mn, Mo, Se, U, V et Zn de sites dans le lac Wollaston touchés par des rejets de la mine du lac Rabbit, située dans le nord-est de la Saskatchewan. Les FE en Cr et en V dans les sédiments sont plus élevés dans le lac Wolf, tandis que le FE en U est plus élevé dans les sédiments superficiels du Petit lac McDonald. Nous n'avons pas pu déterminer la biodisponibilité du Cr, de l'U et du V pour le meunier noir et le grand brochet en raison d'un manque de tissus. Dans les sites touchés de la baie Hidden du lac Wollaston, les éléments As, Cd, Cu, Mn, Mo, Se, et U présentent les FE les plus élevés. Nous n'avons pas analysé de poissons du lac Wollaston. Nous recommandons la réalisation de recherches supplémentaires pour la mine du lac Rabbit en fonction des questions posées dans l'ETIMA.

Mots-clés : exploitation minière de l'uranium; métallothionéine; métaux; poisson; sédiments.

## INTRODUCTION

Over the past decade uranium mines and mills in northern Saskatchewan annually produced about 30% of the world's uranium supply. During this time ore bodies discovered in this area have been and continue to be brought into production. Improved practices for the management and treatment of tailings and effluents are also being implemented to minimize the release of contaminants into receiving waters.

In terms of developing environmental monitoring programs for uranium mines, research on radionuclides may be over-emphasized in importance, and study of other contaminants has been under-emphasized (Saskatchewan Research Council, 1991; Ripley et al., 1996). The regulation of effluent discharges from uranium mines and mills into receiving waters will include the application of an Environmental Effects Monitoring (EEM) program (Environment Canada, 1996, Dumaresq et al., 2002) under the revised Metal Mining Effluent Regulations (MMER). The objective of the metal mining EEM program is to evaluate the effects of mine effluents on the aquatic environment, specifically fishes, fish habitat, and the use of fisheries resources (Environment Canada, 1999; Queen's Printer for Canada, 2001). During the development of the EEM program (Dumaresq et al., 2002), an Aquatic Effects Technology Evaluation (AETE) program (AETE, 1999) was undertaken to evaluate and define meaningful and cost-effective aquatic monitoring tools to be considered for inclusion in the EEM program. Field monitoring tools considered by the AETE program addressed at least one of the following Guidance Questions:

1. Are contaminants getting into the system?
2. Are contaminants bioavailable?
3. Is there a measurable biological response?
4. Are the contaminants in the system causing the observed response?

The objective of this paper is to present information relevant to these Guidance Questions as it pertains to uranium mining and milling in northern Saskatchewan. Fishes and sediments were collected from lakes impacted by effluent discharges from the Key Lake operation which has been ongoing for over twenty years. Fishes were also collected from un-impacted, reference lakes near new mines (the McArthur River and Cigar Lake operations) recently brought into operation or expected in the near future to come into operation. The information in this paper may be included within the EEM structure as part of Site Characterization studies, and provides useful baseline data for related investigations.

This paper consists of a main body and an Appendix. The main body of this report presents information on concentrations of Al, As, Cd, Co, Cu, Fe, Hg, Pb, Mn, Mo, Ni, Se and Zn in sediments, and, with the exception of Al, in fishes from those lakes. Results are reported on metal concentrations in sections of sediment cores and in livers and kidneys of white suckers (*Catostomus commersoni*), northern pike (*Esox lucius*) and lake whitefish (*Coregonus* *clupeaformis*) from those lakes and on metallothionein concentrations in those tissues. In a strict sense, the term “metalloids” can be used for describing trace elements, such as As and Se; however, throughout this discussion the conventional use of the term “metal” (Manahan, 1992; Rainbow, 1993) will be used to describe all the contaminants. The Appendix of this report presents results on concentrations of Cr, U, and V, in sections of sediment cores obtained from lakes at the Key Lake site. Concentrations Al, As, Ba, Cd, Co, Cr, Cu, Fe, Mn, Mo, Pb, Se, U, V, and Zn in sections of cores of sediments impacted in Hidden Bay of Wollaston Lake near the Rabbit Lake operation in northeastern Saskatchewan are also presented in the Appendix.

## MATERIALS AND METHODS

### KEY LAKE SITE

The Key Lake mine is located in north-central Saskatchewan (57° 11’N, 105° 34’W) approximately 70 kilometres east-southeast of Cree Lake (Fig. 1). Open pit mining of the Gaertner ore body commenced in 1982 and was completed in 1987. Open pit mining of the Deilmann ore body commenced in 1986 and was completed in 1997. Subsequent to 1999, stockpiled ore from the Deilmann pit was blended with ore from the McArthur River mine which is trucked to the Key Lake mine for milling. All of the McArthur River ore is transported by truck to the Key Lake mine for milling. From 1994 to 1999, the Key Lake mine was licensed to produce 4.4 million kg of uranium annually. In November, 1999, the licensed annual production limit was raised to 7.4 million kg of uranium to accommodate ore from the McArthur River mine.

Mining and milling operations at Key Lake influence two separate drainages (Fig. 1 Key Lake Project Area); the mill effluent pathway, which consists of the David Creek drainage, and the mine dewatering pathway, which consists of the McDonald Lake-Outlet Creek drainage. Both of these drainages feed into the Wheeler River. Treated mill effluent is discharged to Wolf Lake which drains to Fox Lake through about 500 metres of boggy wetland, and on to Yak Creek, David Creek and then to the Wheeler River. Water from about 50 dewatering wells for the Gaertner and Deilmann open pits discharges into Horsefly Lake, which drains to Little McDonald Lake, McDonald Lake and on to McDonald Creek, Wilson Lake, Outlet Creek and the Wheeler River. High nickel loading associated with the de-watering well discharges led to the construction of a reverse osmosis plant to lower nickel concentrations in the waters discharging to Horsefly Lake. Volumes discharged in 1994 were approximately 1,737,000 $m^3$ of treated mill effluent into the mill effluent pathway and about 10,171,000 $m^3$ of water into the mine dewatering pathway.

Characteristics of lakes in the study area may be summarized as follows:

**Mill Effluent Pathway**

| Lake | Area (ha) | Mean depth (m) | Max. depth (m) | Volume ($m^3$) |
|---|---|---|---|---|
| Wolf | 9.06 | 0.55 | 1.40 | 50, 189 |
| Fox | 14.6 | 1.86 | 4.25 | 271, 192 |

**Mine Dewatering Pathway**

| Lake | Area (ha) | Mean depth (m) | Max. depth (m) | Volume ($m^3$) |
|---|---|---|---|---|
| Horsefly | 6.8 | 7.2 | 15 | 387, 747 |
| Little McD.* | 37.0 | 11.5 | 23 | 4,281,000 |
| McDonald | 326 | 7.8 | 19 | 25,586,000 |

* = Little McDonald Lake

Characteristics of nearby lakes used as Reference Lakes[a] for providing white suckers, northern pike and lake whitefish for this study:

**Key Lake area**

| Lake | Area (ha) | Mean depth (m) | Max. depth (m) | Volume ($m^3$) |
|---|---|---|---|---|
| Zimmer | 347 | 5.8 | 21 | 20,261,000 |

**McArthur River area**

| Lake | Area (ha) | Mean depth (m) | Max. depth (m) | Volume ($m^3$) |
|---|---|---|---|---|
| Toby | 24 | 2.6 | 3.7 | 622,000 |
| Lower Read | 38 | 4.2 | 6.6 | 1,580,000 |
| Boomerang | 25 | 1.0 | 1.8 | 241,000 |
| Little Yalo* | 70 | 5.3 | 9.8 | 3,740,000 |

* = Little Yalowega

**Cigar Lake area**

| Lake | Area (ha) | Mean depth (m) | Max. depth (m) | Volume ($m^3$) |
|---|---|---|---|---|
| Bizarre | 44 | 8.7 | 21 | 3,800,000 |

[a] = Sediment samples were not obtained from these lakes.

The latitude and longitude coordinates for the lakes are:

| Lake Name | Latitude | Longitude |
|---|---|---|
| David[1] | 57° 12’ 00” N | 105° 44’ 30” W |
| Fox[1] | 57° 13’ 19” N | 105° 40’ 51” W |
| Zimmer[1] | 57° 09’ 00” N | 105° 46’ 00” W |
| Little McDonald[1] | 57° 11’ 17” N | 105° 37’ 22” W |
| McDonald[1] | 57° 12’ 00” N | 105° 35’ 00” W |
| Boomerang[2] | 57° 44’ 30” N | 105° 05’ 00” W |
| Lower Read[2] | 57° 44’ 51” N | 105° 06’ 53” W |
| Little Yalowega[2] | 57° 46’ 00” N | 104° 58’ 30” W |
| Toby[2] | 57° 45’ 16” N | 105° 04’ 39” W |
| Bizarre[3] | 58° 04’ 30” N | 104° 35’ 00” W |

[1] Lake in the vicinity of the Key Lake mine.
[2] Lake in the vicinity of the McArthur River mine.
[3] Lake in the vicinity of the Cigar Lake mine.

**SEDIMENTS**

Sediment cores, 5 cm in diameter, were obtained in August 1994 by SCUBA divers and sectioned in a manner similar to that described by Harrison and Klaverkamp (1990). Maintaining sediment surface integrity was achieved by visual observation by the SCUBA diver at the time of obtaining each core. Cores were taken at sites from the deepest part of Wolf Lake, Fox Lake and the northwest bay of Little McDonald Lake. Water depths at these sites were 1.5 metres (Wolf), 4.3 m (Fox), and 17.0 m (Little McDonald). Four cores were obtained from each site. Cores were sliced from top to bottom on site into 1-cm sections to a depth of 10 cm, and into 2-cm sections to a depth of 20 cm or deeper (see Results) immediately following collection. Individual sections were placed in labeled plastic (Whirlpak®) bags, air was excluded, and sections were immediately frozen on dry ice and transported to the Freshwater Institute (FWI), Winnipeg, MB. Sediment samples were prepared by lyophilization at –50° C for three to four days using a Labconco Lyph-lock 12 freeze-dryer.

In the Environmental Chemistry Laboratory of the FWI, individual sections of each sediment core were digested with nitric, perchloric and hydrofluoric acids for the analysis of Al, Cd, Cu, Co, Cr, Fe, Mn, Mo, Ni, Pb, Zn, V and U (Sturgeon et al., 1982). Digests were analyzed for metals and trace elements using a Varian SpectrAA-20 flame atomic absorption spectrophotometer (AAS) equipped with a deuterium continuum, simultaneous background corrector, a Spectrametrics Spectraspan III Direct Current Plasma Emission Spectrometer, and a Hitachi Z-8200 Series Polarized Zeeman Atomic Absorption Spectrophotometer.

For determination of As and Se, sediments were digested with nitric, sulfuric, and perchloric acids, diluted to volume in a 30% hydrochloric acid matrix and analyzed using a semi-automated hydride generating system (Vijan and Wood, 1974) coupled to a Varian SpectrAA-20 AAS as previously described (Harrison et al., 1989; Harrison and Klaverkamp, 1990).

For the determination of Hg, sediment samples were digested in aqua-regia (Dow Chemical Method CAS-AM-70.13) and analyzed using a semi-automated manifold system (Armstrong and Uthe, 1971) coupled to a LDC Model 3200 Mercury Monitor for detection by cold vapour atomic absorption spectrometry.

Certified reference materials were included in the analyses of sediments to evaluate analytical accuracy. Marine sediment standard reference materials (BCSS-1, MESS-1 and PACS-1) certified and distributed by the National Research Council of Canada were used. Results were all within the accepted 95% confidence limits of the certified values.

Means of the metal concentrations in each section from the same depth were calculated from the cores sampled from each sampling site. Data (μg/g, dry wt.) were expressed as depth profiles of mean metal concentrations in the sediments, along with the associated standard errors. Surface-to-background ratios (“Enrichment Factors”) of these concentrations were calculated as the ratio of the concentration in the top 1 cm section to the concentration in the deepest section (> 20 cm for metals in Wolf, Fox and Little McDonald Lake).

**FISHES**

Using gill nets, biologists from FWI and Terrestrial & Aquatic Environmental Managers Ltd. (TAEM), Saskatoon, Saskatchewan captured white suckers, northern pike and lake whitefish during August and September, 1994. FWI personnel captured fishes from Wolf, Fox and Little McDonald lakes. TAEM personnel captured fishes from Zimmer, Boomerang, Little Yalowega, Lower Read, Toby and Bizarre lakes. In the fall of 1997, additional northern pike were captured from Boomerang Lake, Toby Lake, Little Yalowega Lake and Lower Read Lake; additional white suckers from Boomerang Lake and Lower Read Lake, and additional lake whitefish from Little Yalowega and Toby Lakes (Conor

Pacific Environmental Technologies Inc., 1999). No attempts were made to capture fishes from Horsefly or McDonald lakes. Gill nets were checked approximately every 30 minutes, and only live fish were dissected for analyses of metals and metallothionein (MT). All fish were dissected on site.

Before dissection, fishes were either (FWI collected) anesthetized using pH-neutralized tricaine methane sulfonate (500 mg/L), or (TAEM collected) were killed by a blow to the head using a blunt object. Fork length, weight, and sex data were obtained, and structures for aging were removed. White suckers, northern pike and lake whitefish collected by FWI personnel were aged using pectoral fin rays (Don MacDonnell, North/South Consultants Inc., Winnipeg, Manitoba). Fish collected by TAEM were aged (J. Hochbaum, SERM, Fisheries Branch) using the cleithrum for northern pike, the sagittal otolith for lake whitefish, and pectoral fin rays for white suckers. Liver and kidney were removed and placed individually in labeled plastic (Whirlpak®) bags, air was excluded, and tissues were immediately frozen on dry ice and transported to the FWI. Tissues were stored at –90°C until analyzed.

Some attempts were made (see Results section) by FWI biologists to capture small forage fish species at the Key Lake sites using baited minnow traps.

Analytical procedures for metals and trace elements in liver and kidney were similar to those described by Harrison and Klaverkamp (1990). For analyses of Cd, Cu, Pb, Fe, Mn, Mo, Ni, Co, and Zn, tissues were digested using a nitric acid dry-down procedure (Malley et al., 1989). Hg was analyzed by cold vapour atomic absorption spectrometry (Armstrong and Uthe, 1971); and As and Se by semi-automated hydride generation atomic absorption spectrometry (Vijan and Wood, 1974). Certified reference materials were also included in the analyses to assess analytical accuracy. Bovine liver (Standard Reference Material 1577), and oyster tissue (Standard Reference Material 1566), certified and distributed by the U. S. National Bureau of Standards, and dogfish muscle and liver standard reference materials (DORM-2 and DOLT-2) certified and distributed by the National Research Council of Canada were included. Results were all within the accepted 95% confidence limits of the certified values.

Metallothionein (MT) was analyzed by a Hg displacement assay (Klaverkamp, et al., 2000).

**DATA ANALYSES**

Concentrations of metals and metallothionein in fish were analyzed by one-way ANOVA employing Dunnett's multiple comparison procedure. Data were analyzed within species and tissue type. Where data deviated significantly from a normal distribution, non-parametric analysis was applied employing the Kruskal-Wallis test. Relationships between metal concentrations and fish meristics were evaluated by linear regression analysis. Concentrations less than the analytical detection limit were estimated as one-half of the detection limit. The level of $\alpha$ was set at 0.05 in all tests. All statistical analyses were performed using SAS version 8.0 (SAS Institute Inc. Cary, NC, USA, 1999).

Comparisons of tissue metal concentrations to similar concentrations in the literature

In order to compare (see Discussion section), metal concentrations measured in this study as wet weight to concentrations measured in previous studies as dry weight, dry weight concentrations were converted to wet weight concentrations by multiplying the dry weight value by 0.2 (Jarvinen and Ankley, 1999). The converted values are indicated by (w.w. calc.).

## RESULTS

### SEDIMENTS

Table 1 presents data on all metal concentrations in the surficial (top) section of sediment cores and the calculated Enrichment Factor (EF) for each metal in sediments from Wolf and Fox lakes and from the northwest bay of Little McDonald Lake. Mo, As and Se exhibited the highest degree of surficial enrichment in lakes (Wolf and Fox) influenced by treated mill effluent; and sediments from the northwest bay of Little McDonald Lake, which is impacted by mine dewatering discharges, had the highest EFs for Co and Ni. (Table 1; Fig. 2). EFs for molybdenum were high in both Wolf Lake (EF = 720) and downstream Fox Lake (EF = 323), which indicates a high degree of mobility of this metal. Enrichment factors for As, Se and Ni in Wolf Lake (259, 33.5 and 6.1, respectively) were similar to those in Fox Lake (276, 25.8 and 5.2, respectively), which indicates that these contaminants are also highly mobile. EFs for Co and Ni in sediments of Little McDonald Lake were 29.5 and 10.3, respectively. Because Al concentrations are lower in the top 6 to 8 cm of sediments from Wolf and Fox lakes (Fig 2), normalizing the data for other contaminants according to Al concentrations as described by Joshi et al., (1989) would result in even higher EFs.

### FISHES

In the Key Lake area, attempts to collect fishes were made by using gill nets and baited minnow traps. Attempts by FWI biologists using gill nets and baited minnow traps to capture fishes from Wolf Lake were not successful. Attempts using baited minnow traps by these biologists to capture small forage fishes from Fox and Little McDonald lakes were also not successful. Sixteen white suckers from Fox Lake; as well as five lake whitefish and seven northern pike from Little McDonald Lake were captured by DFO personnel using gill nets during the later part of August, 1994. In the fall of 1994 personnel from TAEM Ltd. captured an additional 4 northern pike each from Little McDonald Lake and Zimmer Lake. TAEM Ltd. biologists also captured an additional five lake whitefish from Little McDonald Lake and four lake whitefish from Zimmer Lake.

For the McArthur River and Cigar Lake areas, TAEM Ltd. biologists provided liver and kidney samples from five northern pike and five white suckers from each of Boomerang, Little Yalowega, Lower Read and Toby lakes (McArthur River Project) and from Bizarre Lake (Cigar Lake Project). These five lakes are not impacted by effluent discharges and are referred to as “Reference Lakes” (RLs). Therefore, there were 5 RLs (Bizarre, Boomerang, Little Yalowega, Lower Read and Toby) for white suckers, 6 RLs (the 5 RLs for white suckers plus Zimmer Lake) for northern pike, and one RL (Zimmer Lake) for lake whitefish.

Table 2 presents information on the fork lengths, weights, ages, and sexes of the white suckers, northern pike, and lake whitefish. As a group, white suckers from all reference lakes were significantly longer ($p < 0.005$) and heavier ($p < 0.005$) than white suckers from Fox Lake; however, ages did not differ significantly. White suckers from Bizarre Lake and Boomerang Lake were longer and heavier compared to those from Fox Lake, while those from Lower Read Lake were only heavier.

When combined, northern pike from all reference lakes were significantly longer ($p = 0.01$), heavier ($p = 0.04$) and older ($p = 0.001$) than those from Little McDonald Lake. Northern pike from Boomerang Lake and Lower Read Lake were significantly longer, heavier, and older compared to those from Little McDonald Lake, while those from Bizarre Lake were only significantly older.

Lake whitefish from the reference lake (Zimmer Lake) did not differ significantly in length and weight from those obtained from Little McDonald Lake. Lake whitefish from Zimmer Lake were marginally ($p = 0.06$) older.

**<u>Metal concentrations</u>**

Table 3 presents equation parameters for linear regression analyses that demonstrate significant ($p < 0.05$) relationships between metal concentrations in livers and kidneys and either fish age or fish weight. Regressions of Hg and Se on fish age were significant in livers of both white suckers and northern pike from reference lakes ($r^2$ ranged from 0.17 to 0.50). These relationships were not significant in systems impacted by mining activities, nor were there any other consistent relationships between fish age and metal concentrations. Regressions of Hg on fish weight were significant in kidney of white suckers and northern pike and in livers of white suckers and lake whitefish from reference lakes. Similar to age, these relationships were not significant in systems impacted by mining activities, nor were there any other consistent relationships between fish weight and metal concentration. The relationship of Hg in liver of lake whitefish with fish weight appears artificially strong ($r^2 = 1.0$) due to a sample size of only three fish.

Relative to concentrations observed in livers and kidneys of white suckers from five RLs, metals in these tissues of white suckers from Fox Lake were the highest in some cases, the lowest in others, and similar in a few cases (Table 4). In hepatic and renal tissues of white suckers from Fox Lake, concentrations of metals were highest for those contaminants that demonstrated the highest Enrichment Factors (EFs) in sediments from that lake. The most pronounced differences between concentrations measured in livers and kidneys of white suckers from Fox Lake and those determined in white suckers from RLs were observed for Mo, Se, As and Ni (Table 4). Relative to contaminant concentrations in livers of white suckers from the RLs, concentrations of Mo at 2.11 μg/g ± 0.15 were 6.0 to 8.8 fold higher, and Se concentrations at 17.9 μg/g ± 0.8 were 12.2 and 23.9 times higher in livers from Fox Lake white suckers (Fig 3). In kidneys, Mo at 3.60 μg/g ± 0.64 and Se at 16.1 μg/g ± 1.2 were from 12.0 to 37.5 times higher and from 19.2 to 32.3 times higher, respectively, in tissues from Fox Lake white suckers (Fig 3). In all cases these differences were statistically significant.

All white suckers from the RLs had liver and kidney As concentrations less than the detection limit (DL, 0.04 μg/g); and, with few exceptions, Ni concentrations were below the DL of 0.06 μg/g in those fish (Table 4). Arsenic concentrations in livers of Fox Lake white suckers, however, were 15.3 times higher than the DL, and approximately one-half of these white suckers had As concentrations higher than the DL in their kidneys. Ni concentrations in liver and kidney of Fox Lake white suckers were about 2 times and 5 times, respectively, higher than the DL.

Kidneys of these fish also contained the highest Cu concentrations of 6.5 μg/g ± 1.1, which was from 2.3 to 5.1 times higher than corresponding concentrations in kidneys of white suckers from RLs (Fig. 3).

Iron, at 99 μg/g ± 7, was also the highest in kidneys of Fox Lake white suckers, however, this concentration was only 1.1 to 1.7 times higher than corresponding concentrations found in white suckers from RLs (Table 4), and was not significantly different.

Livers and kidneys of Fox Lake white suckers also contained the lowest concentrations relative to those in white suckers from RLs of some metal contaminants (Table 4). In livers Cu, Zn, Cd, Mn and Hg were lowest, and in kidney Zn, Mn and Cd were generally the lowest. In liver, Hg was from 33% to 70% of concentrations observed in white suckers from RLs. Corresponding percentages for Cu were from 45% to 64% (Fig. 3), Zn ranged from 77% to 98%, and Mn ranged from 48% to 79%. Cd was detectable at concentrations ranging from 0.04 μg/g to 0.16 μg/g in livers of white suckers from RLs, but was less than the DL of 0.02 μg/g in 44% of the Fox Lake white suckers. In kidney, Zn concentrations ranged from 77% to 93% of concentrations observed in white suckers from RLs, and corresponding values for Mn were 19% to 50%. Kidneys of white suckers from 4 of the 5 RLs had Cd concentrations ranging from 0.17 μg/g to 0.62 μg/g. Kidneys from approximately 30% of the white suckers from

Fox Lake and one of the RLs had Cd concentrations less than the DL of 0.02 μg/g.

Consistent differences were not observed between Fox Lake and RLs in concentrations of some metals in livers or kidneys of white suckers (Table 4). Iron in liver demonstrated wide lake-to-lake variability, ranging from 119 μg/g ± 31 to 559 μg/g ± 135, with a value of 259 μg/g ± 20 for Fox Lake white suckers. Intermediate concentrations of Hg and Co were also observed in kidneys from Fox Lake white suckers. Pb concentrations were below the DL of 0.10 μg/g in liver and kidney of white suckers from all lakes. Co was below the DL of 0.05 μg/g in livers of white suckers from all lakes.

Metal concentrations in liver and kidney of northern pike from Little McDonald Lake were the highest observed in some cases and similar in other cases relative to concentrations in northern pike from the six RLs (Table 5). Unlike observations on white suckers, however, no metal was lowest in concentration in northern pike from Little McDonald Lake. In those northern pike, concentrations of Ni, Co and Cd in liver were higher than similar concentrations found in northern pike from the six RLs (Fig 4). Ni at 0.72 μg/g ± 0.12 was at least 12 times higher than concentrations found in liver of northern pike from the RLs. Corresponding values for Co at 0.47 μg/g ± 0.08 were at least 9.5 times higher, and for Cd at 0.31 μg/g were from 2 to 19.3 times higher. It is noteworthy that Co and Ni demonstrated the highest EFs, 29.5 for Co and 10.3 for Ni, in sediments from Little McDonald Lake, whereas Cd did not show surficial enrichment (Table 1).

Concentrations of other metals were highest, but to a lesser degree, in livers of northern pike from Little McDonald Lake. Mo at 0.30 μg/g ± 0.04 was 1.5 to 2.0 times higher than those concentrations observed in northern pike from RLs, Hg at 0.27 μg/g ± 0.11 was 1.3 to 2.8 times higher, Mn at 2.16 μg/g ± 0.37 was 1.3 to 2.8 times higher, and Se at 2.42 μg/g ± 0.12 was 20% to 80% higher. Comparisons to metal concentrations in sediments from Little McDonald Lake demonstrate that Mo and Mn had EFs of 3.1 and 8.8, respectively (Table 1). Se did not demonstrate surficial enrichment, and the EF for Hg was less than 2.

Kidneys of northern pike from Little McDonald Lake also contained the highest concentrations of Ni, Co and Cd (Fig. 4). Ni at 2.55 μg/g ± 0.30 was 27 to 42.5 times higher than corresponding concentrations in kidneys of northern pike from the six reference lakes. Co at 1.89 μg/g ± 0.20 was from 13 to 17 times higher; and Cd at 0.60 μg/g ± 0.09 was from 4 to 15 times higher.

Similar to the case with liver, Hg, Mn, Cu and Mo were also the highest, but to a lesser degree, in kidney of northern pike from Little McDonald Lake (Table 5). Hg at 0.34 μg/g ± 0.14 was 1.6 to 3.8 times higher; Mn at 1.38 μg/g ±

0.34 was 1.3 to 3.1 times higher; Cu at 2.05 μg/g ± 0.18 was 1.6 to 2.8 times higher; and Mo at 0.17 μg/g ± 0.04 was 1.5 to 2.5 times higher.

Similar concentrations were observed for the other metals in livers or kidneys of northern pike from Little McDonald Lake and RLs. As was the case for white suckers, Fe in northern pike liver showed wide lake-to-lake variability. Concentrations ranged from 69 μg/g ± 24 to 1,038 μg/g ± 900 with northern pike from Little McDonald Lake having a concentration of 317 μg/g ± 91. Concentrations of Zn in liver and kidney, Cu in liver, and Se in kidney, were intermediate in northern pike from Little McDonald Lake and the RLs. With rare exceptions, Pb concentrations were below the DL of 0.10 μg/g in liver and kidney of all northern pike. Arsenic was below the DL of 0.40 μg/g in both tissues for all northern pike analyzed.

Notable differences between metal concentrations in liver and kidney of lake whitefish from Little McDonald Lake and Zimmer Lake were observed (Table 6). Ni concentrations in liver of 0.57 μg/g ± 0.12 and in kidney of 4.66 μg/g ± 0.72 in lake whitefish from Little McDonald Lake were 9.4 and 7.2 times higher, respectively, than corresponding concentrations in lake whitefish from Zimmer Lake. Co concentrations of 0.23 μg/g ± 0.05 observed in livers of Little McDonald Lake lake whitefish were 3.4 times higher than those in Zimmer Lake lake whitefish. Other differences were less pronounced and included 42% higher Cu in kidney and 40% higher Se and 34% higher Mo than the concentrations observed in livers of lake whitefish from Zimmer Lake. Lower concentrations of Hg and Fe in kidneys and livers of lake whitefish from Little McDonald Lake were also observed with values 21% to 46% of concentrations in Zimmer Lake fish.

Interactions between metals were evaluated by regression analyses. Cu concentrations were significantly correlated with Mo concentrations in several instances. This relationship was strongest ($r^2$ = 0.74) in kidneys of white suckers from Fox Lake (Fig. 5, Left panel). Weak, but significant, correlations between Cu and Mo were also observed in kidneys ($r^2$ = 0.22) and livers ($r^2$ = 0.24) of white suckers from reference lakes and in livers ($r^2$ = 0.25) of northern pike from reference lakes. Hg concentrations were significantly correlated to Se in kidneys of white suckers from Fox Lake; in livers and kidneys of white suckers from the reference lakes; and in livers of northern pike from the reference lakes. In comparisons between Hg and Se, the strongest correlation ($r^2$ =0.62) was observed in kidneys of Fox Lake white suckers (Fig. 5, lower panel), and the weakest ($r^2$ = 0.34) in kidneys of white suckers from the reference lakes. Hg concentrations in livers of white suckers from Fox Lake showed little relationship with Se concentrations ($p$ = 0.43; $r^2$ = 0.05).

**<u>Metallothionein concentrations</u>**

Concentrations of MT in livers and kidneys exhibited species-, tissue- and lake-to-lake variabilities. MT concentrations in livers of Fox Lake white suckers were significantly lower than those observed in livers of white suckers from all RLs;

however, MT concentrations in kidneys of Fox lake white suckers were in the middle of the range (Fig. 6). Concentrations in livers of Fox Lake white suckers were 153 μg/g ± 19, which represents 39% to 59% of the concentrations found in livers of white suckers from RLs. Concentrations of MT in kidneys of Fox Lake white suckers were 53 μg/g ± 7. Corresponding mean concentrations in kidneys of white suckers from two of the reference lakes (Bizarre and Little Yalowega) were 60 and 64 μg/g; and for white suckers from three of the RLs (Lower Read, Boomerang and Toby) were 25, 33 and 39 μg/g.

MT concentrations in livers and kidneys of northern pike from Little McDonald Lake were in the middle of ranges observed in tissues of northern pike from RLs (Table 7). Concentrations in livers and kidneys of Little McDonald Lake northern pike were 264 μg/g ± 30 and 109 μg/g ± 9, respectively. Northern pike from four RLs had higher mean concentrations with a range of 325 μg/g to 548 μg/g in their livers and northern pike from two RLs had lower mean concentrations of 158 μg/g and 226 μg/g (Table 7). MT in kidneys of northern pike from reference lakes had a range of mean concentrations of 82 μg/g to 168 μg/g.

MT concentrations at 71 μg/g ± 17 in kidneys of Little McDonald Lake lake whitefish were not significantly different from those seen in Zimmer Lake lake whitefish (Table 6). MT concentrations at 96 μg/g ± 17 in livers of Little McDonald Lake lake whitefish, however, were only 53% of those observed in lake whitefish from Zimmer Lake.

Regression analyses were conducted for MT against Cu and against Cu+Cd+Zn. Strong associations between MT and Cu concentrations were observed in livers of white suckers from Fox Lake ($r^2$ = 0.80; Fig. 7A) and RLs ($r^2$ = 0.89; Fig. 7B), of northern pike from RLs ($r^2$ = 0.84; Fig. 7C), and of lake whitefish from Little McDonald Lake ($r^2$ = 0.88; Fig. 7D). Regressing MT concentrations against Cu+Cd+Zn provided little change in $r^2$ and p-values in livers of white suckers from reference lakes and of northern pike from reference lakes. By adding Cd and Zn, however, the association was weakened (change in $r^2$ from 0.80 to 0.55) for liver MT in white suckers from Fox Lake, and eliminated ($r^2$ from 0.88 to 0.27) for MT in livers of lake whitefish from Little McDonald Lake. Adding Cd and Zn to Cu, however, resulted in a significant ($p < 0.03$), but weak ($r^2$ = 0.28), association with MT concentrations in kidneys of white suckers from Fox Lake and reference lakes. Fish age was not a determinant of hepatic and renal MT concentrations.

## Discussion

Concern for adverse effects arising from uranium mining and milling activities in northern Saskatchewan on fishes and their habitats has largely been focussed on pathways and effects of uranium and radionuclides (Bernstein and Swanson, 1989; Ripley et al., 1996). Although other metals have received less attention, knowledge about their presence, bioavailability and adverse effects is required to develop appropriate field biomonitoring programs.

Sediments serve as sinks for metals, and are important as a route of uptake for these contaminants into aquatic biota (Ingersoll et al., 1997). Metals in sediments can be directly taken up by organisms that live in or near the sediments, and these organisms can mediate contaminant transfer to the water column or to other trophic levels (Rosenberg et al., 1997).

Relationships between concentrations of metals in sediments and aquatic biota are highly complex and involve a diverse array of abiotic and biotic factors (Luoma and Fisher, 1997). A wide variety of operationally-defined methods have been developed for predicting the bio-availability of an individual metal or of simple mixtures of metals bound to sediments (Luoma and Fisher, 1997). For example, acid volatile sulfide appears to be the dominant sediment-binding phase for Cd, Cu, Ni, Pb and Zn (Swartz and DiToro, 1997). No single method is available, however, for predicting the bioavailability of all sediment-bound, metals to fishes. Applying the variety of operationally-defined methods for predicting the bioavailability of metals in sediments to fishes was beyond the scope of this study.

Sediments are not static repositories of metals (Tessier et al., 1994). Complex chemical and biological processes can influence the vertical distribution of metals and, therefore, can affect the interpretation of sediment core profiles in quantifying the magnitude and/or history of metal contamination by anthropogenic activities (Johnson, 1998). For redox active elements, such as As, Fe and Mn, enrichment factor calculations may result in an over or underestimate of recent inputs as a result of redox processes.

The sediment sampling program conducted by the consulting firm (TAEM) in 1997 on Boomerang Lake, Little Yalowega Lake, Lower Read Lake and Toby Lake provided pre-operational data for a suite of inorganic contaminants (Conor Pacific Environmental Technologies Inc., 1999). Although these data were not peer-reviewed and published in a primary scientific journal, they form a basis for comparison to the results from this study. With some exceptions, contaminant concentrations in surficial sediments from these four reference lakes were comparable to concentrations found in deeper (> 20 cm) sediment sections from Wolf Lake, Fox Lake, and Little McDonald Lake. Exceptions included generally lower concentrations of Al, As, Fe, Ni and U, and generally higher concentrations of Mn, in the sediments from RLs. Surficial sediments from RLs and deeper sediments from impacted lakes should reflect baseline conditions in both cases. The exceptions, therefore, likely represent fundamental geological differences between lake sediments. Because Se accumulated to concentrations of concern in fish from Fox Lake (see below), it is unfortunate that analyses for Se were not conducted on the 1997 sediment samples obtained from the reference lakes.

Water samples were not collected in late August of 1994 from impacted lakes at the Key Lake site or from the reference lakes in August and September of 1997. In late August of 1997, Pyle et al. (2001) collected water samples from Fox Lake, Little McDonald Lake, and David Lake, a reference lake near the Key Lake site. David Lake flows into David Creek, which receives the flow from Yak Creek, the receiving water immediately downstream of Fox Lake. Mine dewatering discharges or treated mill effluent do not influence David Lake. This lake is 139 ha in surface area, 1.85 m in mean depth, 3.5 m in maximum depth and 2.58 x 106$m^3$ in volume. The water samples analyzed by Pyle et al. (2001) were collected at the same time of the year as the sediment and fish samples. Because there were no changes in effluent treatment practices during that time interval, results from Pyle's study are relevant to this study. Pyle's results for water quality parameters and for total and dissolved metal concentrations in water are summarized in Tables 8 and 9, respectively.

Relative to the reference lake (David Lake), the waters of Fox Lake are acidic, hard, and high in conductivity, TDS, carbon dioxide and turbidity (Table 8). The differences are close to two orders of magnitude in the cases of hardness, conductivity and TDS. Little McDonald Lake water is approximately 3 fold higher in hardness, conductivity and TDS than the water of David Lake. Little McDonald Lake is less turbid than David Lake.

Concentrations of total metals and the percentages dissolved are presented in Table 9. Fox Lake water has the highest concentrations of As, Cd, Cu, Fe, Mn, Mo, Se and Hg. Little McDonald Lake water contains the highest concentrations of Co, Ni and Zn. At the Key Lake mining site, high Mo concentrations and high Ni concentrations characterize uranium mill effluents and dewatering effluents, respectively (Pyle et al., 2001). For some of the metals there is considerable variation in the percentage of total that is dissolved. For reasons presented above, Pyle's data are relevant to this study because fish from impacted lakes accumulate metals from water and dietary routes of uptake (Farag et al., 1995; Handy, 1996; Bird et al., 1998). The data summarized in Table 9 are thoroughly discussed by Pyle et al. (2001), and, with the exception of Cd, will not be considered further in this Discussion.

## CATEGORIES OF METALS AND TRACE ELEMENTS, AND SELECTION OF TISSUES

There are two general categories of metals and metalloids, essential and non-essential (Simkiss and Taylor, 1995). By meeting diverse nutritional, metabolic and respiratory functions, essential metals are required for good health and reproductive competence. These chemical elements generally function as co-factors in metallo-enzymes and other metallo-proteins. Tissue concentrations of essential metals are generally regulated by transport systems in cellular and sub-cellular membranes (Simkiss, 1998; Brown and Depledge, 1998). Excess amounts of essential metals cause harmful effects, and deficiency states can also exist when adequate stores are not available. Tissue concentrations of non-essential metals are generally not regulated, although these chemical elements can interfere and compete with essential metals in membrane transport systems (Simkiss, 1998; Chan, 1998). Non-essential metals are not required for biological functions, and are of concern when present in amounts that exceed their thresholds of toxicity.

Essential metals measured in fish tissues in this study include Mo, Se, Co, Fe, Cu, Mn and Zn (Sorensen, 1991). As, Pb, Cd, Ni and Hg are generally regarded as non-essential metals. Ni may be an essential metal in birds and mammals, because Ni deficiency states have been observed in those animals (Nielsen, 1987; Keen, 1996). Arsenic may be essential for the offspring of pregnant rats (Keen, 1996; Chan, 1998).

Unfortunately, there is a lack of knowledge on what constitutes “normal concentrations” of essential metals in white suckers, northern pike and lake whitefish; on their requirements for essential metals; on their toxicity thresholds (tissue residues) and on their mechanisms of toxic action. Therefore, interpreting the biological and toxicological consequences of concentrations of essential and non-essential metals in renal and hepatic tissues of these fish species is difficult. Understanding linkages between concentrations of metals in aquatic habitats to concentrations accumulating in specific tissues of aquatic organisms to concentrations producing adverse effects in those organisms is needed for developing environmental fate models, environmental risk assessments and environmental risk management strategies (McCarty and Mackay, 1993).

Because of the knowledge gaps for white suckers, northern pike and lake whitefish, this discussion will focus on comparing metal concentrations in hepatic and renal tissues of those fish species collected from lakes in the Key Lake area to other studies that:

1. relate concentrations in tissues to harmful effects, observed mostly in laboratory-based exposures, in other freshwater fish species;
2. measure concentrations in renal and hepatic tissues from natural populations of other freshwater fish species, or;
3. in the cases of Cu, Zn, Cd and Hg; analyzed these metals in hepatic and renal tissues from natural populations of white suckers, northern pike and lake whitefish.

This third category is unique from the other two because, as discussed in the sections on those four metals, larger data sets exist for concentrations of those metals in livers and kidneys of the three fish species.

With the exception of Hg, which exists in fish largely as lipid-soluble, methyl mercury, the use of fish muscle is a poor choice of tissue for determining the bio-availability of metals (Forstner and Wittmann, 1981; Olsson et al., 1998). It has long been documented that organs, such as liver and kidney, from fish exposed in the laboratory to high concentrations of metals contain higher, dose-related concentrations than skeletal muscle (Reed et al., 1968; Benoit et al., 1976; Holcombe et al., 1976; Holcombe et al., 1979). Studies on natural populations of fishes from metal-contaminated habitats have also confirmed that, with the exception of mercury (Heit and Klusek, 1985; Parks et al., 1991), muscle is a poor choice for determining metal availability. Because skeletal muscle usually comprises over 55% of a fishes' total body weight, the use of whole-body analyses (Johnson, 1987) is also not a good choice because "muscle-dilution" of metals that are primarily distributed to other organs, e.g. liver and kidney, occurs. Liver and kidney have been shown to be appropriate choices (Bradley and Morris, 1986; Harrison and Klaverkamp, 1990; Munkittrick et al., 1991; Miller et al., 1992). Liver and/or kidney are also critical target organs for the harmful effects produced by many of these metals (Sorensen, 1991; Chang, 1996).

**<u>Molybdenum and Selenium</u>**

Concentrations of Mo were higher, especially in livers and kidneys of white suckers from Fox Lake than those concentrations measured in white suckers from RLs (Table 4). Mean concentrations of Mo observed in livers and kidneys of white suckers from Fox Lake were from about 6 to 38 times higher than those observed in tissues of white suckers from RLs. The mean concentrations measured in Fox Lake white suckers are also an order of magnitude higher than those observed in previous studies of free-swimming fishes from contaminated habitats. Kokanee salmon (*Oncorhynchus nerka*) from a deep, oligotrophic reservoir near a Mo mining area in Colorado contained mean concentrations of 0.100 μg/g and 0.124 μg/g in their livers and kidneys, respectively; and rainbow trout from the same reservoir contained 0.233 μg/g and 0.147 μg/g, respectively (Ward, 1973). Rainbow trout (*Oncorhynchus mykiss*) held for two weeks in cages located 1.6 km from a Mo tailings pile contained 8.6 μg/g (w.w. calc.) and 5.2 μg/g (w.w. calc.) in their livers and kidneys, respectively (Kienholz, 1977). No overt adverse effects were noted for these trout. Additional research is recommended on the early life history stages of the white sucker population of Fox Lake, because embryo and larval stages of fishes are over 3 orders of magnitude more sensitive to Mo than the juvenile stage (Eisler, 1989).

Selenium is of special concern because it readily accumulates and causes harmful effects in freshwater fish (Sorensen, 1991; Lemly, 1996). Adverse consequences of Se impacts on natural fish populations have been documented (Lemly, 1985; Lemly and Smith, 1987). In a Se-contaminated ecosystem in California, Se concentrations increased from water to sediments to aquatic plants and invertebrates to fish (Saiki and Lowe, 1987). Selenium toxicity was observed in bluegills (*Lepomis macrochirus*) that consumed Se-contaminated benthic macro-invertebrates from Belews Lake in North Carolina (Finley, 1985). In a whole-lake study using metal radio-isotopes at the Experimental Lakes Area (ELA) in northwestern Ontario, $^{75}Se$ was in a group of metals, including $^{203}Hg$ and $^{65}Zn$, that demonstrated the highest Concentration Ratios of individual metal radio-isotope activities in fish relative to those activities in water (Harrison et al., 1990). After 63 days, $^{75}Se$ activities in fathead minnows (*Pimephales promelas*) were 7,660-fold higher than activities in epilimnetic water; and, after 352 days, activities in lake trout (*Salvelinus namaycush*) were 14,000-fold higher than those found in the whole lake water column.

Causes of concern for Se toxicity in natural fish populations include reproductive problems and teratogenic deformities in larval offspring from sexually mature and reproducing adult fish with elevated Se body burdens (Lemly, 1996; Lemly, 1997). A toxic effects threshold for the overall health and reproductive vigor of freshwater and anadromous fish of 3 μg Se/g (w.w. calc.) in livers has been defined (Lemly, 1996). Direct maternal transfer of Se to progeny is the major vector for accumulation in embryos, and the cause of edema and lordosis in developing larval fish (Schultz and Hermanutz, 1990). Se concentrations of 4 to 8 μg/g (w.w. calc.) in visceral tissues, including livers and kidneys, from reproducing adults has been proposed as confirming a diagnosis of Se-induced teratogenesis in the offspring (Lemly, 1997).

Se concentrations in livers and kidneys of white suckers from Fox Lake are approximately 2 to 4.5 times higher than this range of concentrations. Concentrations in livers of these fish are about 6 times higher than the toxic effects threshold of 3 μg Se/g (w.w. calc.; Lemly, 1996). Mean concentrations of Se in liver and kidney of 17.9 μg/g and 16.1 μg/g, respectively, in white suckers from Fox Lake are among the highest observed in natural populations of freshwater fish (Lemly, 1996). Biomonitoring programs at Se-contaminated sites in the United States have demonstrated hepatic concentrations ranging from 6.7 μg/g to 34 μg/g (Sorensen, 1991). In these fish, livers had the highest Se concentrations followed by ovaries and skeletal muscle.

Comparing Se concentrations observed in laboratory studies of other fish species to those measured in white suckers from Fox Lake also indicates that harmful effects may be occurring in the Fox Lake population. Mean concentrations in livers of 10.2 μg/g (w.w. calc.) in rainbow trout were correlated with changes in hematological parameters (Hodson et al., 1980). Hepatic concentrations of 5.8 μg/g (w.w. calc.) and 6.8 μg/g (w.w. calc.) in bluegills were correlated with reproductive failure (Hermanutz et al., 1992) and mortality (Finley 1985), respectively. Rainbow trout fed Se-contaminated diets at 11.4 μg/g for 16 weeks in the laboratory accumulated 51.3 μg/g and 11.2 μg/g in liver and kidney, respectively (Hicks et al., 1984). These fish exhibited increased Ca and Mg concentrations in their kidneys, renal lesions and body weight decreases.

For comparisons to other natural white sucker populations, livers of white suckers from 5 lakes impacted by atmospheric emissions from a base-metal smelter and from 3 reference lakes contained Se concentrations of 2.2 μg/g ± 0.3 and 0.9 μg/g ± 0.1, respectively (Harrison and Klaverkamp, 1990). Mean Se concentrations in livers of white suckers from three reference lakes in the ELA ranged from 1.11 to 2.45 μg/g (Harrison and Klaverkamp, 1990). White suckers from Keg Lake, a reference lake near the community of Red Lake, Ontario, contain 1.04 μg/g ± 0.07 and 0.71 μg/g ± 0.03 in their livers and kidneys, respectively (Klaverkamp et al., 2002).

Although adult fish from natural populations may appear to be healthy, continued monitoring of the Fox Lake white sucker population is recommended because it could disappear due to major adverse impacts on the early life-history stages. These adverse impacts could occur because of high concentrations of Se; Mo; a combination of the two; or other factors. It is noteworthy that white suckers from Fox Lake were the same age, but were shorter and weighed less than white suckers combined from the RLs (Table 2). To better understand relationships between metal contamination and effects on Fox Lake white suckers, migratory patterns of these fish should also be investigated.

Seventy to ninety percent mortality occurred in larval fathead minnows (<24h old) exposed *in situ* for 7 days in Fox Lake (Pyle et al., 2001). Although Mo and As concentrations in water were strongly correlated with mortality, the authors concluded that these metals were not responsible based largely on published 96h $LC_{50}$ data. Concentrations of Se in water were also strongly correlated with mortality, and arguments are presented based on the Se literature for dietary Se being the causative factor (Pyle et al., 2001). The authors, however, ignore literature on the protective effects of As on Se toxicity. High As concentrations were observed in water (Table 9), sediments (Table 1) and in livers and kidneys of white suckers from Fox Lake (Table 4). It has long been documented (Dubois et al., 1940; Levander, 1977) that As exposure (waterborne or diet-borne) protects against dietary Se toxicity in mammals and birds. Recent studies also present evidence for arsenic's protective effects against Se toxicity in larval razorback sucker (*Xyrauchen texanus*) larvae (Hamilton et al., 2002). The studies by Pyle et al. (2001), were also confounded by high mortality (mean of 20%) in the controls, and in exposures affected by high winds and forceful wave action.

With a few exceptions, mean concentrations of Se and Mo were also higher in livers and kidneys of northern pike and lake whitefish from Little McDonald Lake than concentrations in similar tissues of northern pike from the six RLs and in lake whitefish from Zimmer Lake (Tables 5 and 6). Livers of northern pike from Little McDonald Lake contained 2.4 μg/g of Se, which was higher than concentrations observed in northern pike from all of the reference lakes. Renal Se concentrations in these fish were the highest compared to reference lakes, with the exception of northern pike from Bizarre Lake, which had slightly higher concentrations of Se in their kidneys. At 0.30 μg/g and 0.17 μg/g, mean molybdenum concentrations in livers and kidneys, respectively, were also highest in northern pike from Little McDonald Lake (Table 5). Concentrations of Se and Mo in livers of lake whitefish from Little McDonald Lake were approximately 40% higher than those found in livers of lake whitefish from Zimmer Lake; whereas concentrations in kidneys were about the same (Table 6). Although these concentrations were the highest, or among the highest, observed in these two fish species, the magnitudes of differences observed between impacted and reference northern pike and lake whitefish populations are considerably less than similar differences between white sucker populations. Northern pike from Little McDonald Lake, however, were generally younger than northern pike from RLs, and, therefore, were exposed to metals for a shorter duration.

Comparisons to other natural populations of northern pike and lake whitefish outside of this study area indicate that concentrations of Se are not exceptionally high in fishes from Little McDonald Lake (Tables 5 and 6). Livers of northern pike from 5 lakes near a base metal smelter and from four reference lakes contained Se concentrations of 2.52 μg/g ± 0.18 and 1.76 μg/g ± 0.06, respectively (Harrison and Klaverkamp, 1990). Livers and kidneys of northern pike from a reference site in Yellowknife Bay of Great Slave Lake contained Se concentrations of 0.92 μg/g ± 0.06 and 1.27 μg/g ± 0.09, respectively (Jackson et al., 1996). Se concentrations in livers and kidneys of lake whitefish from the same reference site in Yellowknife Bay were 1.40 μg/g ± 0.04 and 0.93 μg/g ± 0.08, respectively. Northern pike from a reference lake (Keg Lake) in northwestern Ontario contained 1.43 μg/g ± 0.10 and 1.09 μg/g ± 0.07, respectively, in their livers and kidneys (Klaverkamp, et al., 2002).

It is surprising, however, that Mo and Se concentrations in livers and kidneys of northern pike from Little McDonald Lake were the highest or among the highest relative to those observed in northern pike from the six reference lakes in northern Saskatchewan. It is surprising because several factors contribute either to the dilution of metal discharges into Little McDonald Lake or to the migration of northern pike within the ecosystem. First, mine dewatering discharges travel through and are diluted by Horsefly Lake (>387,000 $m^3$) before reaching Little McDonald Lake. Second, in comparing the situation to Fox Lake and its white sucker population, Little McDonald Lake (>4,000,000 $m^3$) by itself is considerably larger, and capable of greater dilution than Fox Lake (<272,000 $m^3$). Third, Little McDonald Lake is directly connected to McDonald Lake (>25,000,000 $m^3$) with the result being that both of these lakes actually comprise one large body of water. Fourth, even though northern pike were caught in the northwest basin of Little McDonald Lake, these fish most likely migrate throughout the entire system. Despite these four factors, the results demonstrate that Se and Mo discharged by uranium mining activities are readily available to fish, even to large-bodied species living in large lakes with capabilities of swimming considerable distances in these water-bodies.

**<u>Arsenic and Nickel</u>**

Relative to white suckers from the five RLs, white suckers from Fox Lake had the highest As and Ni concentrations in their livers and kidneys (Table 4). It is difficult to quantify the differences, however, because concentrations of these two contaminants were below DLs in most livers and kidneys of white suckers from the RLs. For example, the mean concentration of 0.61 μg As/g in livers of white suckers from Fox Lake was over 15 times the detection limit (DL; 0.04 µg/g); whereas similar concentrations in livers of white suckers from RLs were all less than this limit. In kidneys of 53 % of the fish from Fox Lake, however, As accumulated to concentrations greater than this DL; whereas in white suckers from the RLs arsenic concentrations in kidneys were less than this DL. At 0.11 µg/g, the mean Ni concentration in livers of white suckers from Fox Lake was approximately twice the DL (0.06 µg/g). Similar analyses in livers of white suckers from 4 of the 5 RLs were less than this limit, and in one reference lake 40% of the white suckers contained less than the limit. In kidneys, however, the differences were greater. A mean Ni concentration of 0.29 µg/g in kidneys of white suckers from Fox Lake was about five times higher than the DL. Similar values for white suckers from the RLs ranged from 60% to 100% of the fish having less than the Ni DL in their kidneys.

Arsenic concentrations in livers and kidneys of northern pike from Little McDonald Lake and the 6 RLs were less than the DL; while Ni concentrations were much higher in tissues of northern pike from Little McDonald Lake relative to northern pike from RLs (Table 5); and in similar tissues from lake whitefish relative to those of fish from Zimmer Lake (Table 6). In northern pike, differences were most pronounced in kidneys (Table 5). The mean Ni concentration of 2.6 µg/g in kidneys of northern pike from Little McDonald Lake was from 27 to about 43 times higher than mean concentrations found in northern pike from the RLs. Ni concentrations in livers of northern pike from the RLs were less than the DL (0.06 µg/g) in five of the six lakes; and in northern pike from the sixth lake the mean concentration was 0.09 µg/g (Table 5). Even this concentration, which is the highest observed in northern pike from RLs, is eight times lower than the mean Ni concentration of 0.72 µg/g observed in livers of northern pike from Little McDonald Lake. In livers and kidneys of lake whitefish from Little McDonald Lake, mean Ni concentrations were 0.57 µg/g and 4.66 µg/g, respectively (Table 6). These concentrations are higher by approximately 9 and 7 times, respectively, than similar concentrations found in lake whitefish from Zimmer Lake.

Comparing As concentrations in livers and kidneys of white suckers from Fox Lake to concentrations reported for natural populations of other freshwater fish species demonstrates that similar concentrations are found in hepatic tissues of fishes from contaminated habitats. For example, brown trout (*Salmo trutta*) from a contaminated section of the Clark Fork River in Montana, USA accumulated significantly higher concentrations of As in their livers (0.32 µg/g; w.w. calc.) and kidneys (2.26 µg/g; w.w. calc.) as compared to brown trout from a reference site (Farag et al., 1995). Arsenic concentrations in sediments at the contaminated site were 102 µg/g, d.w. (Brumbaugh et al., 1994)). Rock bass (*Ambloplites rupestris*) from Moira Lake, which has concentrations of As in sediments of 1,000 µg/g, d.w., accumulated 0.21 µg/g in their livers (Azcue and Dixon, 1994). Concentrations of As in livers of brown trout from the Clark Fork River and Moira Lake and from Fox Lake are in the same order of magnitude, but

As concentrations in kidneys of Fox Lake white suckers are an order of magnitude less than those observed in the brown trout.

Hepatic As concentrations in white suckers from Fox Lake, brown trout from the Clark Fork River and rock bass from Moira Lake are within the same order of magnitude as those causing cellular and sub-cellular damage in lake whitefish. Hepatic As concentrations as low as 0.3 μg/g caused nuclear, architectural and structural alterations, areas of inflammation and focal necrosis in lake whitefish consuming food with an arsenic concentration of 10 μg/g (Pedlar et al., 2002a). In lake whitefish consuming dietary As at 100 μg As per g food for 30 days, mean As concentrations of 4.4 μg/g and 0.76 μg/g in liver and kidney, respectively, were observed (Pedlar and Klaverkamp, 2002). These elevated As concentrations were associated with decreased growth, increased hepatic MT concentrations, and histopathological lesions in liver and gall bladder (Pedlar et al., 2002a).

Lake trout (*Salvelinus namaycush*) that were fed As contaminated diets accumulated mean As concentrations of about 1.5 μg/g and 1.0 μg/g in their livers and kidneys, respectively (Pedlar et al., 2002b). These trout had significant increases in concentrations of plasma lipid peroxides and significant decreases in liver somatic indices and weight gain. Macroscopic lesions of the gall bladder were observed in the lake trout exposed to arsenic. These lesions were characterized by hemorrhaging, diffuse whitening of the gall bladder wall and a thickened appearance (Pedlar et al., 2002b).

There are few studies on As concentrations in livers and kidneys of freshwater fish from non-impacted, reference habitats. In a study that analyzed livers of white suckers and northern pike from lakes contaminated by base-metals, but not As, in the area of Flin Flon, Manitoba (Harrison and Klaverkamp, 1990), As concentrations were less than the DL of 0.05 μg As/g in that study. Bohn and Fallis (1978) observed concentrations of 0.14 μg As/g in liver of landlocked arctic char (*Salvelinus alpinus*) in the vicinity of a lead/zinc mine.

Northern pike, white suckers and lake whitefish from reference sites in the geographical vicinity of active gold mining appear to have higher baseline As concentrations. Northern pike from a reference site in Yellowknife Bay of Great Slave Lake had 0.17 μg As/g ± 0.01 and 0.21 μg As/g ± 0.02 in their livers and kidneys, respectively (Jackson et al., 1996). Similarly, lake whitefish from the same site had 0.13 μg As/g ± 0.01 and 0.19 μg As/g/g ± 0.03 in their livers and kidneys, respectively. In livers and kidneys of northern pike from Keg Lake, Ontario, As concentrations were 0.13 μg As/g ± 0.02 and 0.26 μg As/g ± 0.03, respectively (Klaverkamp et al., 2002). White suckers from this lake contained 0.17 μg As/g ± 0.01 and 0.30 μg As/g ± 0.02 in their livers and kidneys, respectively.

Hepatic and renal Ni concentrations in northern pike and lake whitefish from Little McDonald Lake and in white suckers from Fox Lake are generally similar to those concentrations found in freshwater fishes from other Ni-contaminated freshwater ecosystems. Common whitefish (*Coregonus lavaretus*) exposed to Ni contaminated sediments (2,227 μg/g; d.w.) from base-metal mining activities in the Kola Peninsula, Russia, accumulated 4.42 μg/g (w.w. calc.) and 0.62 μg/g (w.w. calc.) in their kidneys and livers, respectively (Kashulin and Reshetnikov, 1995). Corresponding mean Ni concentrations in kidneys and livers of northern pike from this site were 1.50 μg/g (w.w. calc.) and 0.24 μg/g (w.w. calc.), respectively. Comparable concentrations were found in similar tissues of arctic charr and brown trout from the same region of Russia (Moiseenko et al., 1995). In livers from the benthic-feeding Prussian carp (*Carassius auratus*) collected from the Danube River in Yugoslavia, Ni concentrations were 0.48 μg/g (Maletin et al., 1996). Rainbow trout from the Augraben River in Italy accumulated 1.2 μg/g and 1.6 μg/g in their livers and kidneys, respectively (Dallinger and Kautzky, 1985). Even higher concentrations ranging from 10.7 to 17.0 μg/g in livers and from 11.8 to 51.6 μg/g in kidneys were observed about 25 years ago in a variety of fish species collected from the Wanapitei River near the mining and smelting activities in the Sudbury area of Ontario (Hutchinson et al., 1976).

In lake whitefish fed Ni-contaminated diets, cellular and sub-cellular alterations were observed at nickel concentrations of 0.3 μg/g in their livers and 3.8 μg/g in their kidneys (Ptashynski and Klaverkamp, 2002; Ptashynski et al., 2002). In livers of lake whitefish exposed to dietary Ni, areas of focal necrosis and altered bile ducts were observed. Histopathological alterations were also observed throughout the posterior kidneys, glomeruli, tubules, collecting ducts and hematopoietic tissue. The frequency of altered distal tubules and fields of view with alterations increased with Ni dose and duration of exposure (Ptashynski et al., 2002).

In lake trout fed Ni-contaminated diets, responses different from controls were observed at tissue concentrations of 1.3 μg Ni/g in their livers and 7 μg Ni/g in their kidneys (Ptashynski et al., 2001). These lake trout had significant increases in renal MT concentrations and plasma lipid peroxide concentrations. Significantly lower concentrations of $K^+$, $Na^+$ and $Cl^-$ in the plasma of these fishes were also reported. The lake trout exposed to Ni also exhibited histopathological lesions in their intestines and significant decreases in weight (Ptashynski et al., 2001).

Few studies have documented hepatic and renal Ni concentrations in natural populations of white suckers, northern pike and lake whitefish. Concentrations of 0.18 μg/g ± 0.02 and 0.68 μg/g ± 0.09 in livers and kidneys, respectively, measured in white suckers from Keg Lake, Ontario are higher than those observed in white suckers from Fox Lake (Table 5). Conversely, northern pike from Little McDonald Lake have hepatic Ni concentrations that are 7.2 and

24 times higher, respectively, than concentrations observed in livers of northern pike from Yellowknife Bay (Jackson et al., 1996) and Keg Lake (Klaverkamp et al., 2002). Similarly, Ni concentrations in kidneys of northern pike from Little McDonald are 11.8 and 23.6 times higher than corresponding concentrations in kidneys of northern pike from Keg Lake (Klaverkamp et al., 2002) and Yellowknife Bay (Jackson et al., 1996). Hepatic and renal Ni concentrations in lake whitefish from Little McDonald Lake are 7 and 19 times higher, respectively, than corresponding concentrations observed in lake whitefish from Yellowknife Bay (Jackson et al., 1996).

**Cobalt, Iron, Manganese and Lead**

Co accumulation in fish also appeared to be related to concentrations in surficial sediments. In Fox Lake this concentration (μg/g, d.w.) was 12.4 μg/g ± 5.6; whereas in Little McDonald Lake sediments the Co concentration (μg/g, d.w.) was 446 μg/g ± 119 μg/g (Table 1). White suckers from Fox Lake demonstrated no pronounced differences in hepatic and renal Co concentrations from Co concentrations measured in livers and kidneys of white suckers from the RLs. Co concentrations, however, were considerably higher in livers and kidneys of fish from Little McDonald Lake. In kidney of northern pike, concentrations were 13 to 17 times higher in northern pike from Little McDonald Lake than those observed in RL northern pike; and in northern pike from Little McDonald Lake, hepatic Co concentrations were approximately 10 times higher than the D.L., whereas all RL fish had concentrations less than the D.L. In lake whitefish from Little McDonald Lake, hepatic Co concentrations were about 3.5 times higher than those observed in fish from the RL.

Early studies on Co in fish demonstrated an affinity for blood and blood-rich organs, particularly kidney and liver (Reed et al., 1968; Reed, 1971; Kimura and Ichikawa, 1972). Carp (*Cyprinus carpio*), which were fed cobalt contaminated food over a 63-day period, accumulated the highest concentrations in kidney (Baudin and Fritsch, 1987). Experimental additions of $^{60}$Co to lakes in northwestern Ontario demonstrated the highest accumulation in kidneys of lake trout (Harrison et al., 1990) and lake whitefish (Bird et al., 1998). Samples from four fish species exposed to effluents from copper-nickel operations in the Kola Subarctic region of Russia were pooled and analyzed (Moiseenko et al., 1995). Mean concentrations of Co in kidneys ranged from 0.32 μg/g to 6.5 μg/g (w.w., calc.), and from 0.22 μg/g to 1.1 μg/g (w.w., calc.) in liver. These concentrations are within the same order of magnitude as those observed in this study.

Iron concentrations in livers of white suckers from Fox Lake and in livers and kidneys of northern pike from Little McDonald Lake were similar to those concentrations observed in white suckers and northern pike from RLs. Mean Fe concentrations in kidneys of white suckers from Fox Lake were highest relative to reference lakes, but to a lesser degree than Mo, Se, Cu and Ni (Table 4). Fox Lake surficial sediments were enriched by a factor of about 3, whereas little, if any, enrichment was observed in Little McDonald Lake (Table 1). There are few reports on hepatic and renal Fe concentrations in white suckers and northern pike for comparison. In white suckers analyzed in this study, mean hepatic Fe concentrations ranged from 119 μg/g to 559 μg/g; whereas the concentration observed in livers of white suckers from a reference lake (Keg Lake) in northwestern Ontario was 106 μg/g ± 19 (Klaverkamp et al., 2002). In northern pike analyzed in this study mean hepatic Fe concentrations ranged from 69 μg/g to 1038 μg/g, and mean renal Fe concentrations ranged from 75 μg/g to 125 μg/g. Hepatic and renal Fe concentrations in northern pike from Keg Lake were 88 μg/g ± 21 and 78 μg/g ± 5, respectively. Wide lake-to-lake variability in hepatic and renal Fe concentrations was also observed in white suckers from eight acidic, slightly acidic or circumneutral lakes in south-central Ontario (Bendell-Young and Harvey 1989). Mean Fe concentrations ranged from 71 μg/g to 223 μg/g (w.w., calc.) in livers and from 69 μg/g to 135 μg/g (w.w., calc.) in kidneys. It is noteworthy that, despite the paucity of data, the highest hepatic and renal concentrations of Fe were observed in white suckers and northern pike from northern Saskatchewan lakes. Fe can initiate severe harmful effects through a process of lipid peroxidation (Bondy, 1996; Nieminen and Lemasters, 1996), hence the trend of higher Fe concentrations in fishes from northern Saskatchewan should be verified through additional studies. Measurement of Fe speciation is also recommended because iron bound to proteins, such as ferritin, may not be as toxic as free iron. To evaluate the toxicological relevance of Fe concentrations these studies could also use indicators of lipid peroxidation in fish (Cooley et al., 2000; Pedlar et al., 2002; Ptashynski et al., 2002).

Mn concentrations in freshwater sediments are generally higher in areas of mining activities (Heiny and Tate, 1997). Compared to other metals, Mn is relatively non-toxic (Agrawal and Srivastava, 1980; Garg et al., 1989; Gonzalez et al., 1990; Stubblefield et al., 1997). The combination of labile, inorganic Mn and acidic pH, however, was proposed as the cause of mortality in fish exposed to snow melt in the streams of central Sweden (Nyberg et al., 1995).

Few studies have measured Mn concentrations in livers and kidneys of freshwater fish. Brown trout exposed to radiolabelled Mn accumulated the highest concentrations in liver with lesser amounts in kidney (Rouleau et al., 1995). Maximum Mn accumulation in the finfish (*Oreochromis mossambicus*) was observed in liver (Ayyadurai et al., 1994). Adult channel catfish (*Ictalurus punctatus*) used in laboratory studies contained a mean concentration of 1.67 μg/g (wet wt.) in their livers (Griffin et al., 1999). This concentration is within the same order of magnitude observed in white suckers and northern pike in this study. No primary publications were found that presented data for Mn concentrations in kidneys of freshwater fish.

With rare exceptions, hepatic and renal lead concentrations were below the detection limit in white suckers and northern pike from impacted and reference lakes. Lead analyses were not conducted on livers and kidneys of lake whitefish. Similarly, concentrations of less than 0.10 μg Pb/g were documented in livers and kidneys of white suckers and northern pike from Keg Lake (Klaverkamp et al., 2002), and in livers of northern pike from rivers in eastern England (Barak and Mason, 1990). White suckers from an acidic lake in the Adirondack Mountain area of New York contained mean concentrations (w.w. calc.) of 0.26 μg/g and 0.18 μg/g in their livers and kidneys, respectively (Stripp et al., 1990). Fishes exposed to effluents discharged into the Subernarekha River at Ghatsila from copper and uranium operations contained mean Pb concentrations (w.w. calc.) of 9.6 μg/g and 4.4 μg/g in their livers and kidneys, respectively (Singh et al., 1990). Hepatic and renal concentrations of Cd and Cu were also very high in these fishes, and their populations are disappearing.

**Copper, Zinc, Cadmium and Mercury**

Relative to most metals discussed above, larger published data sets exist for Cu, Zn, Cd and Hg in livers and kidneys of white suckers, northern pike and lake whitefish (Tables 10, 11 and 12). In Canada, interest in these four metals is due to a large number of base-metal mines, a wide geographical area having base-metal mining activities, and the substantive role of base-metal smelters in discharging sulfur dioxide, Hg and the other metals to the atmosphere (Environment Canada, 1991 and 1996; Pierce et al., 1998).

Relative to concentrations in fish from reference lakes in the area, copper concentrations were lowest in livers of Fox Lake white suckers (Table 4) and of lake whitefish from Little McDonald Lake (Table 6). In reports by other investigators, hepatic Cu concentrations in white suckers (Table 10) and lake whitefish (Table 12) were also generally higher than those observed in fish from contaminated lakes in this study. Surprising exceptions were observations on white suckers from lakes impacted by acidic precipitation in Adirondack Park in New York state (Stripp et al., 1990), and in some lakes in south-central Ontario (Bendell-Young and Harvey, 1989). Additional research is required to determine whether white suckers in affected lakes (Table 10) are under stress from inadequate Cu stores. Focussed research should also determine whether the associated decrease in MT concentrations (see below) results in fish that are more vulnerable to harmful effects produced by Cd, Cu, Hg and/or Zn (Roesijadi, 1992; Mason and Jenkins, 1995; Olsson et al., 1998).

In kidneys of the three fish species sampled from Fox Lake or Little McDonald Lake, copper concentrations were higher than those corresponding observations in all fish from RLs (Tables 4, 5 and 6). Renal copper concentrations in white suckers from other contaminated lakes were similar to those observed in Fox Lake white suckers (Table 10). Average Cu concentrations in kidneys of white suckers from contaminated systems were approximately three times higher than corresponding concentrations in white suckers from RLs. Northern pike from reference sites in Canada contain about one-half the copper concentration found in northern pike from Little McDonald Lake (Table 11). Kidneys of northern pike from contaminated systems in the Kola peninsula in the sub-arctic region of Russia contained lower mean concentrations of Cu than those measured in northern pike from a reference lake (Table 11). The authors speculated that the exceptionally high concentrations of Ni in these fish interacted with copper to produce lower concentrations (Kashulin and Reshetnikov, 1995). Kidneys of lake whitefish from Little McDonald Lake contained higher Cu concentrations than those measured in the reference lake (Zimmer Lake), but concentrations were lower than corresponding concentrations observed in common whitefish from contaminated and reference systems in sub-arctic Russia (Table 12). Similar to the situation with northern pike, the authors state that high Ni concentrations in contaminated fish result in Cu concentrations lower than those observed in reference fish (Kashulin and Reshetnikov, 1995).

Like copper, zinc concentrations in livers and kidneys of white suckers from Fox Lake were also lower than corresponding concentrations in white suckers from the RLs in this study (Table 4). Zinc concentrations in white suckers from Fox Lake were also at the low end of the range documented by other investigators (Table 10). Some of the differences, especially Zn concentrations in liver, were not pronounced; and, in several cases, Zn concentrations were higher in white suckers from reference systems (Table 10) than those observed in Fox Lake white suckers. Zn concentrations in renal and hepatic tissues of northern pike and lake whitefish from Little McDonald lake were similar to those observed in reference lakes in this study (Tables 5 and 6) and in other reference and contaminated systems (Tables 11 and 12).

Hepatic and renal tissues of white suckers from Fox Lake also contained the lowest Cd concentrations observed in this study (Table 4); and their concentrations were also lower than those measured in other investigations (Table 10). In this study, Cd concentrations in livers and kidneys of northern pike from Little McDonald Lake were from 2 to 19 times higher than those measured in northern pike from reference lakes (Table 5). This is surprising because there was no surficial enrichment (E.F. < 1.0) of Cd in sediments of Little McDonald Lake (Table 1); and because low Cd concentrations were measured by Pyle et al. (2001) in the waters of this lake (Table 9). Northern pike collected from lakes near the Flin Flon smelter and from RLs in northern Saskatchewan contained similar hepatic Cd concentrations to those in Little McDonald Lake (Table 11). Other investigations of northern pike from reference systems document hepatic and renal Cd concentrations ranging from about 10% to 50% of those measured in Little McDonald Lake (Table 11).

In this study, Hg concentrations in livers of white suckers were also lowest in fish from Fox Lake (Table 4). Renal Hg concentrations in these white suckers were not different from those observed in white suckers from RLs. When compared to studies of white suckers from other Canadian lakes, however, no consistent differences in Hg concentrations were observed in the Fox Lake fish (Table 10). With one exception, northern pike from Little McDonald Lake contained the highest concentrations of Hg in their livers and kidneys (Tables 5 and 11). The exception consists of hepatic Hg concentrations in northern pike from lakes in northeastern Saskatchewan that were used as reference systems for studies on the impacts of the Flin Flon smelter (Harrison and Klaverkamp, 1990). Hepatic concentrations of selenium, which is known to lower hepatic Hg concentrations in freshwater fishes (Jackson, 1998), were also the lowest in those northern pike.

There have been few studies on Cd and Hg concentrations in livers and kidneys of lake whitefish. Cd concentrations in tissues of lake whitefish from Little McDonald Lake were lower than those observed in lake whitefish from Zimmer Lake, but higher than those in lake whitefish from Yellowknife Bay of Great Slave Lake (Table 9).

**Comparisons between fish caught in 1994 and 1997**

Additional fishes collected and analyzed in 1997 included northern pike from Boomerang Lake, Toby Lake, Little Yalowega Lake, and Lower Read Lake; white suckers from Boomerang Lake and Lower Read Lake; and lake whitefish from Little Yalowega and Toby lakes (Conor Pacific Environmental Technologies, 1999). Coefficients of variation, especially for Cu, Fe and Mo in livers of northern pike collected in 1997 were relatively large; frequently extending to over 100%. No consistent significant differences were observed between metal concentrations in livers and kidneys of the northern pike and the white suckers collected in 1994 and 1997.

In lake whitefish captured in 1997 from Little Yalowega Lake and Toby Lake, concentrations of Cd and Mo in liver and of Cd, Co, Mo and Ni in kidney were significantly less than concentrations found in similar tissues of lake whitefish collected in 1994 from Zimmer Lake and Little McDonald Lake. Information was not provided on length, weight, age and sex for fish collected and analyzed in 1997, however, so variations in contaminant concentrations may also be due to differences in these parameters.

**Metal Interactions**

Because sediments were contaminated to a considerable degree by As, Mo, Ni and Se in Fox Lake, and by Co, Mn, Mo and Ni in Little McDonald Lake, it was not surprising to observe elevated concentrations of these metals in the hepatic and renal tissues of fishes from those lakes. Contamination of receiving waters and resident organisms by metals discharged from uranium mining and milling operations has been documented by others (Saskatchewan Research Council, 1991; Ripley et al., 1996; Conor Pacific Environmental Technologies Inc., 1999; Hynes, 1990).

It was surprising to discover that relative to concentrations found in white suckers from RLs in this study, the lowest concentrations of Cu, Cd, Zn, Hg and Mn were present in white suckers from Fox Lake. In these fish each of these five elements was lowest in livers, and Zn, Cd and Mn were also lowest in kidneys. Similar results were not observed in northern pike from Little McDonald Lake.

Interactions between metals may be responsible for the observations on white suckers from Fox Lake. In general, when considering detoxification effects, metal interactions have been described as being the rule rather than the exception (Yu, 2001). For example, Mo is known to lower tissue Cu concentrations in mammals (Eisler, 1989). In mammals, Mo toxicity, which includes weight loss, severe diarrhea, early abortions, early neo-natal deaths and parental death, results from Cu deficiency states (Keen, 1996). The mechanism of this interaction involves the formation of thiomolybdates and molybdoproteins that chelate Cu and prevent the absorption of Cu (Eisler, 1989; Simkiss and Taylor, 1995; Keen, 1996). Zn deficiencies are also caused by high tissue concentrations of Mo in mammals (Parada, 1981). The underlying mechanism is also likely due to interference by thiomolybdates with Zn at the same receptor sites used for copper absorption (Simkiss and Taylor, 1995). High tissue Ni concentrations, such as those measured in this study, may also be responsible for lowering Cu and Zn concentrations. Common whitefish and northern pike from a lake with Ni-contaminated sediments had lower concentrations of Cu and Zn in their livers and kidneys than fish from an uncontaminated lake (Kashulin and Reshetnikov, 1995). Mn is known to be transported through Ca channels that are also involved in diffusion processes through cell membranes by other divalent cations, e.g. Co, Mg and Cd (Simkiss and Taylor, 1995). Competition at these channels by divalent cations may help to explain the low Mn concentrations in livers and kidneys of white suckers from Fox Lake. Selenium's action in lowering Hg concentrations in muscles and livers of freshwater fishes is well documented (see review by Jackson, 1998). The positive correlation observed between Hg and Se in the kidneys of white suckers from Fox Lake (Fig. 5) may be related to a mechanism involving increased renal excretion of Hg by Se. These interactions as well as other metal-metal interactions, which have largely been observed in mammals (Landis and Yu, 1999; Yu, 2001), require additional research in fishes.

**<u>Metallothionein</u>**

Metallothionein is a ubiquitous, low-molecular-weight (7 kDa), heat-stable, cysteine-rich, heat shock protein, whose structure is remarkably similar across taxa (Kille et al., 1992). Under non-pathological physiological conditions, the primary function of this protein is believed to be its participation in the maintenance of copper and zinc homeostasis (Olsson, 1996). Because MT scavenges free radicals which may be generated in the presence of metals, MT also functions as an antioxidant (Mason and Jenkins, 1995; Olsson et al., 1998). MT synthesis is induced by trace amounts of non-essential metals, Cd and Hg, and by excessive levels of Cu and Zn in fish (Roesijadi, 1992; Wood et al., 1996). In turn, MT sequesters these metals, thus functioning as a detoxification protein (Roesijadi, 1992; Mason and Jenkins, 1995).

MT induction in tissues of fishes inhabiting metal-contaminated habitats has received a great deal of attention over the past decade and more. MT has been advocated as a reliable biomarker of metal exposure and an early-warning bio-indicator of effects (Benson et al., 1990; Klaverkamp et al., 1991; Olsson, 1996; Klaverkamp et al., 1997; Olsson et al., 1998; Klaverkamp et al., 2000). The use of MT in fish and macro-invertebrates as a tool in field biomonitoring programs has been the subject of reviews (Benson et al., 1990; Roesijadi, 1992; Mason and Jenkins, 1995; Olsson, 1996; Couillard, 1997).

MT was selected in this study as a measurable endpoint in relation to the third AETE Guidance Question, “Is there a measurable biological response?”. Because of its role in regulating and detoxifying metals, MT induction is also relevant to the fourth AETE Guidance Question, “Are the contaminants in the system causing the observed response?”. Because of its sensitivity and ecological relevance in protecting fish and macro-invertebrates from metal toxicity, MT was the only biochemical response selected for evaluation by the AQUAMIN (Environment Canada, 1996) and AETE (Couillard, 1997) exercises.

In this study, conclusions on metal and MT concentrations in livers and kidneys of lake whitefish are not made, because only four lake whitefish were captured from one reference lake. Lake whitefish from additional reference lakes, preferably four or more, are required before conclusions can be made on metals and MT in lake whitefish from Little McDonald Lake and on relationships between metals and MT in this species. This requirement for lake whitefish from additional reference lakes is especially crucial in light of recent evidence demonstrating high concentrations of metals in surface sediments from Zimmer Lake relative to corresponding concentrations in David Lake (Golder Associates Ltd., 2002). Therefore, differences in metal and MT concentrations in tissues of lake whitefish from Little McDonald Lake and of lake whitefish from reference lakes may be even more pronounced than those presented in this report.

There were no significant differences between renal MT concentrations measured in white suckers and northern pike from impacted lakes and those concentrations measured in fish from associated RLs. Renal MT concentrations in white suckers from Fox and RLs were weakly, but significantly, correlated with only the sum of renal Cu, Zn, and Cd concentrations. Similar regression analyses for renal MT against Cu, Zn or Cd, and their sum, in northern pike demonstrated no significant correlations. It is surprising, however, that renal MT concentrations in northern pike from Little McDonald Lake were not significantly different from concentrations observed in northern pike from RLs, because concentrations of Cu, Cd and Hg were the highest in kidneys of northern pike from Little McDonald Lake. Although concentrations of Zn and Cd in kidneys of white suckers from Fox Lake were the lowest measured, concentrations of Cu were about 2 to 5 times higher than renal Cu concentrations in white suckers from RLs. The lack of significant differences between renal MT concentrations measured in white suckers from Fox Lake and those observed in white suckers from RLs may be due to a balance of effects between low Zn and Cd (therefore, low MT induction) and high Cu (induction of MT).

Cu was the primary determinate of MT in livers of white suckers from Fox Lake and associated RLs; northern pike from RLs; and lake whitefish from Little McDonald Lake. Adding Cd and Zn to Cu in regression analyses provided no significant changes in coefficients of correlation and statistical significance for MT concentrations in livers of white suckers and northern pike from RLs. Hepatic Cu concentrations in livers of northern pike from Little McDonald Lake were not different from those concentrations in northern pike from RLs. It is not surprising; therefore, that hepatic MT concentrations in northern pike from Little McDonald Lake are not different from those measured in northern pike from RLs. Similarly, because livers of white suckers from Fox Lake contained the lowest Cu concentrations, it is not surprising that Fox Lake white suckers had significantly lower hepatic MT concentrations. Because Cu, Zn, Cd and Hg are known to induce MT and because hepatic concentrations of MT were also the lowest, the low metal concentrations likely contributed to the low induction of hepatic MT in white suckers from Fox Lake. The cause of low hepatic Cu, Zn, Cd and Hg concentrations may be due to the interactions between those metals and Mo, Ni, Mn, Se and/or other metals as described above.

**Summary of Results**

In this section, a brief summary of results is presented as responses to the Guidance Questions developed by the AETE program (AETE, 1999) for consideration in developing the metal mining EEM program:

A. “Are contaminants getting into the system?”

Analyses for Al, As, Cd, Co, Cu, Fe, Hg, Pb, Mn, Mo, Ni, Se and Zn in sections of sediment cores obtained from lakes at the Key Lake uranium mining site demonstrated, through the calculation of Enrichment Factors (EFs), contamination originating from mine-related discharges.

1. The greatest degree of contamination (EFs > 6) in lakes receiving treated mill effluents was observed for Mo, As, Se, Fe and Ni. A high degree of metal mobility in affected lakes was observed for Mo, As, Se, and Ni. In those lakes, the following metals demonstrated EFs ranging from 1.7 to 4.8: Co, Cu, Hg, and Pb.
2. In lakes receiving mine dewatering discharges, the greatest degree of contamination was observed for Co, Ni and Mn with EFs ranging from about 9 to 30. In these lakes, EFs ranging between 2.5 and 3.9 were observed for As, Pb, Mo, and Zn.

Note: As presented in Pyle et al., (2001), the waters of Fox Lake and Little McDonald Lake also contained high concentrations of metals (Table 9).

B. “Are contaminants bioavailable?”

The bioavailabilities of these metals were determined by analyzing livers and kidneys of white suckers, northern pike and lake whitefish from impacted lakes. Concentrations of these metals in hepatic and renal tissues were compared to similar measurements made on white suckers from 5 reference lakes (RLs), northern pike from 6 RLs, and lake whitefish from 1 RL. Only white suckers could be captured from Fox Lake, a downstream receptor of treated mill effluent; whereas only northern pike and lake whitefish were collected from Little McDonald Lake, a downstream receptor of mine dewatering discharges. Metals were bioavailable to fishes as demonstrated by the following observations:

1. Molybdenum. Hepatic and renal tissues of white suckers from the impacted Fox Lake contained 6 to about 38 times higher Mo concentrations than those measured in tissues of white suckers from RLs. Similar tissues of northern pike from the impacted Little McDonald Lake contained 1.5 to 2.5 times higher Mo concentrations than those observed in northern pike from the RLs.
2. Arsenic. Hepatic As concentrations in white suckers from Fox Lake were over 15 times higher than the detection limit of 0.04 µg/g; whereas concentrations in livers of white suckers from RLs were less than this detection limit.
3. Selenium. Hepatic and renal Se concentrations in white suckers from Fox Lake were from 12 to 32 times higher than those measured in similar tissues of white suckers from RLs. Livers of northern pike from Little McDonald Lake contained 1.2 to 1.8 times higher Se concentrations than those observed in livers of northern pike from RLs.
4. Nickel. Hepatic and renal Ni concentrations in white suckers from Fox Lake were at least 2 to 5 times higher than those observed in white suckers from RLs. Renal Ni concentrations in northern pike from Little McDonald Lake were from 27 to about 42 times higher than those measured in kidneys of northern pike from RLs. In lake whitefish from Little McDonald Lake, hepatic and renal Ni concentrations were from 7 to 9 times higher than those measured in livers and kidneys of lake whitefish from Zimmer Lake.
5. Manganese. Mn concentrations were highest in hepatic and renal tissues of northern pike from Little McDonald Lake; whereas concentrations in hepatic and renal tissues of white suckers from Fox Lake were the lowest observed.
6. Copper. Concentrations of Cu were highest in kidneys of white suckers from Fox Lake and of northern pike from Little McDonald Lake. Hepatic Cu concentrations in white suckers from Fox Lake, however, ranged from 45% to 64% of concentrations measured in white suckers from RLs.
7. Cadmium. Hepatic and renal Cd concentrations in northern pike from Little McDonald Lake were from 2 to 19 times higher than those measured in northern pike from RLs. Similar tissues from white suckers in Fox Lake had the lowest Cd concentrations.
8. Cobalt. Relative to concentrations measured in northern pike tissues from the RLs, Co concentrations were at least 9.5 times higher in livers, and from 13 to 17 times higher in kidneys, of northern pike from Little McDonald Lake. Hepatic Co concentrations in lake whitefish from Little McDonald Lake were about 3.5 times higher than those observed in lake whitefish from Zimmer Lake.

9. Mercury. Hepatic and renal Hg concentrations were the highest in northern pike from Little McDonald Lake.
10. Manganese. Hepatic and renal Mn concentrations were also the highest in northern pike from Little McDonald Lake.
11. Iron, Cobalt and Lead. Hepatic and renal concentrations of Fe, Co and Pb in white suckers from Fox Lake, and of Fe and Pb in northern pike from Little McDonald Lake, were similar to those observed in tissues of fishes from their respective RLs.
12. Hepatic Cu, Zn, Cd, Hg and Mn concentrations and renal Zn, Cd and Mn concentrations in white suckers from Fox Lake were lower than similar concentrations in white suckers from all the RLs.

C. "Is there a measurable biological response?"

Responses to these metals were evaluated by measuring whole-body parameters of length, weight, and age; relative to concentrations of Cu, Zn, Cd and Hg, by quantifying hepatic and renal metallothionein (MT) concentrations.

1. White suckers from Fox Lake were not different in age from the white suckers combined from the RLs, but the white suckers from Fox Lake were shorter and lighter in weight.
2. Northern pike from Little McDonald Lake were generally younger and, therefore, exposed for a shorter duration than northern pike from the RLs.
3. Strong, positive correlations between hepatic MT and Cu concentrations were observed in white suckers from Fox Lake, white suckers from reference lakes, northern pike from reference lakes, and lake whitefish from Little McDonald Lake.
4. No differences were observed in renal MT concentrations between white suckers from Fox Lake and renal MT concentrations measured in white suckers from RLs. Hepatic MT concentrations in white suckers from Fox Lake, however, were the lowest measured.
5. No differences were observed in hepatic and renal metallothionein (MT) concentrations between northern pike from Little McDonald Lake and those concentrations measured in northern pike from RLs.

**Recommendations for research**

1. To further address questions on the magnitude and extent of contamination of aquatic ecosystems, water and sediments should be collected at the same time period and analyzed for metals.
2. Analyses of sediment cores, using 1 cm sections to the 10 cm depth and 2 cm sections for depths > 10 cm, obtained from the deepest part of lakes should be the method of choice for determining the magnitude and extent of sediment contamination. Because of wide variations in concentrations of metals in background sediments, surface-to-background ratios (Enrichment Factors, EFs) provide an indication of anthropogenic inputs (see Discussion). These EFs can be useful in assessing temporal changes within individual lakes and in making lake-to-lake spatial comparisons. Sediment cores, and subsequent determinations of EFs, from the RLs should be obtained,

sectioned and analyzed in order to establish better relationships between metal concentrations in sediments and those concentrations measured in livers and kidneys of fishes from those lakes.

3. To address questions pertaining to bioavailabilities of metal contaminants to fishes, liver and kidney of large-bodied fish species should be the tissues of choice. The use of muscle is appropriate only for Hg. Additional tissues, such as bone, gills, gonads and scales should also be evaluated for their use in determining metal bioavailability.
4. Additional efforts should be made to capture small-bodied fish species, such as lake chub (*Couesius plumbeus*) and slimy sculpin (*Cottus cognatus*), and analyze their total visceral contents as an alternative or complement to the use of large-bodied species. A research project that compares results obtained using these two general categories of fishes is needed to assist the selection of fish species in field biomonitoring programs.
5. The role of metal contamination in producing smaller white suckers in Fox Lake should be investigated further. As a starting point, a research project that evaluates indicators of lipid peroxidation as well as branchial, gonadal, hepatic and renal histopathologies in these white suckers and in white suckers from the RLs should be undertaken. Alternative approaches, such as investigating food types and diversities, and their availabilities and abundances should also be undertaken.
6. To address questions pertaining to whether white suckers, northern pike and lake whitefish are responding to metals, research is required on these fish species to determine whether concentrations of essential metals are normal or whether they exceed toxic thresholds. Because concentrations of Mo and Se were higher than those producing adverse effects in other studies, research in this area is urgently required into Mo and Se effects on early life history stages and reproductive competence of adults.
7. Research is also required to determine toxic thresholds for non-essential metals in white suckers, northern pike and lake whitefish.
8. Investigations are needed into whether metal-metal interactions are influencing the accumulation, distribution and toxicology of metals. Starting points for this research could include the potential for effects of Mo on Cu, Cd, Hg and Zn; of Ni on Cu and Zn; and of Se on Hg, Cu, Cd and Zn, and of Se and As interactions.
9. The consequences of low hepatic MT in white suckers from Fox Lake should be evaluated to examine whether those fish are more vulnerable to Cd, Hg, Cu and Zn.

## ACKNOWLEDGEMENTS

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North Environmental Services Limited Partnership assisted with the preparation of Figure 1. Financial support for the study was made available through the DFO Toxic Chemicals Program and the Fish Habitat Management Program.

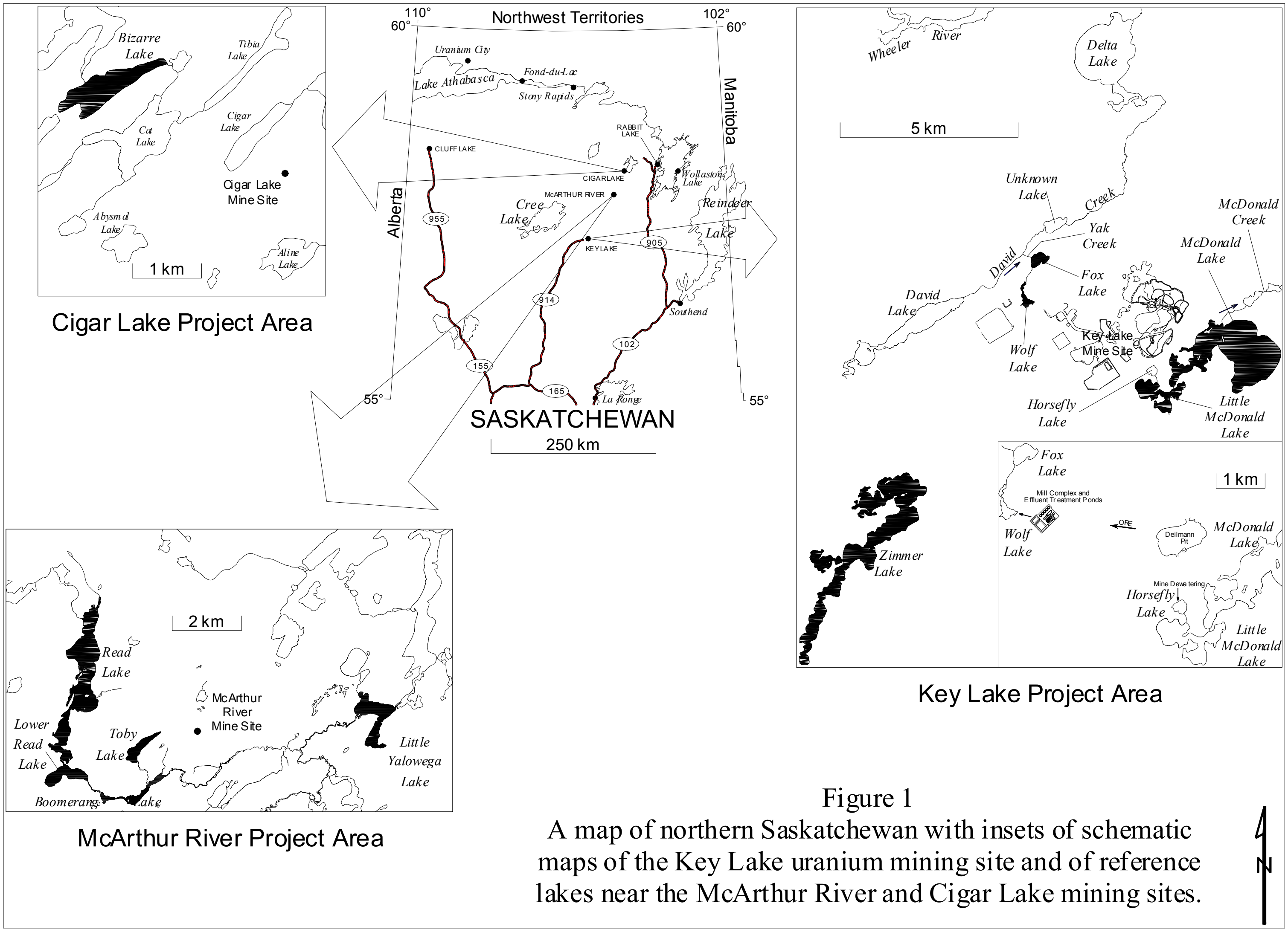


Figure 1
A map of northern Saskatchewan with insets of schematic maps of the Key Lake uranium mining site and of reference lakes near the McArthur River and Cigar Lake mining sites.

**Figure 2.** Sediment core profiles of Mo, As, Se, Ni, Co, and Al for lakes affected by (a.) mining and milling effluents: Wolf (∃) and Fox (■) Lakes; (b.) mine dewatering discharges: Little McDonald Lake (●). Data are expressed as mean (μg/g dry wt.) ± S.E.M.; n = 4.

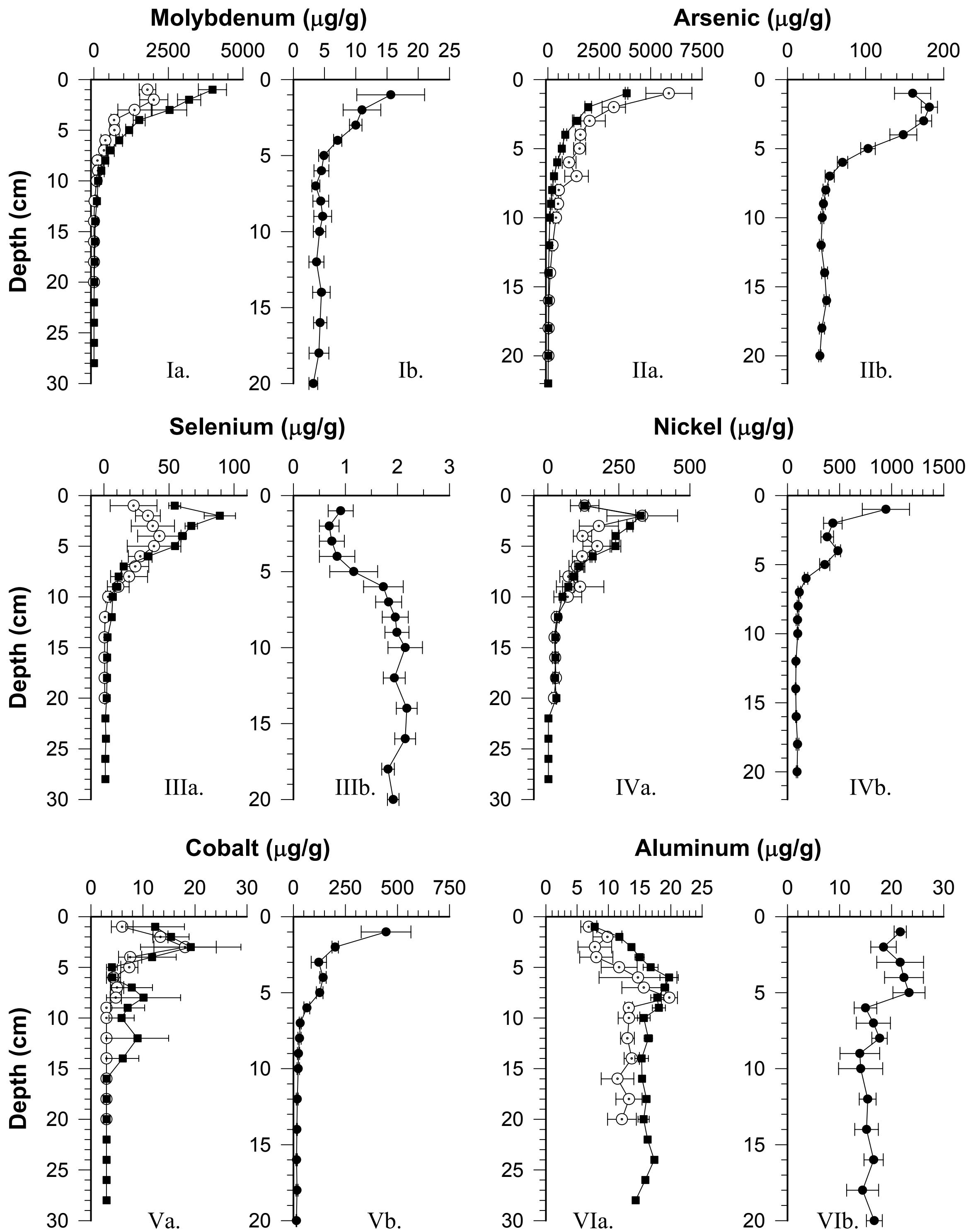

**Figure 3.** Concentrations (μg/g, wet wt.) of Mo, Se and Cu in livers and kidneys of white suckers from Fox Lake (F) and five reference lakes (B = Bizarre Lake, G = Boomerang Lake, R = Lower Read Lake, Y = Little Yalowega Lake and T = Toby Lake). Data are expressed as mean ± S.E.M. See Table 2 for n values.

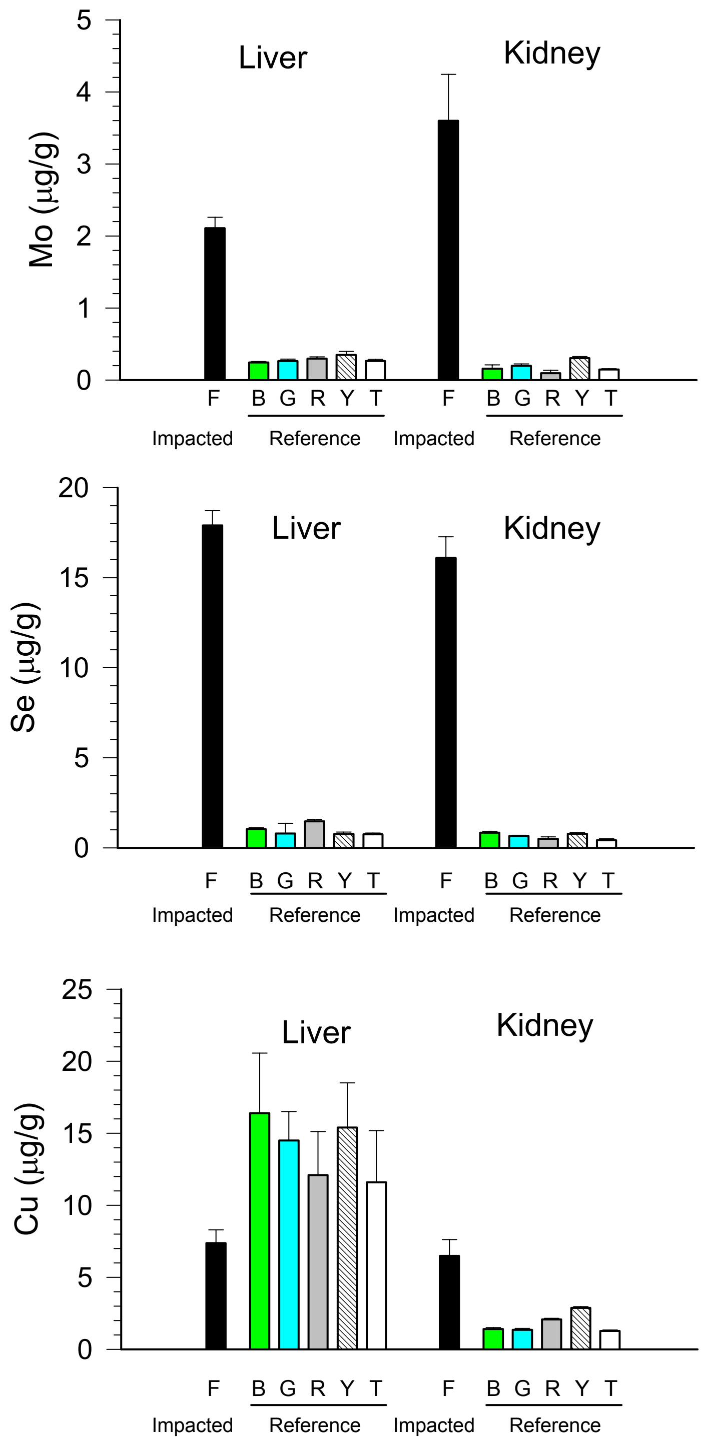

**Figure 4.** Concentrations (μg/g, wet wt.) of Ni, Co and Cd in livers and kidneys of northern pike from Little McDonald Lake (L) and six reference lakes (Z = Zimmer Lake, B = Bizarre Lake, G = Boomerang Lake, R = Lower Read Lake, Y = Little Yalowega Lake and T = Toby Lake). Data are expressed as mean ± S.E.M. See Table 2 for n values.

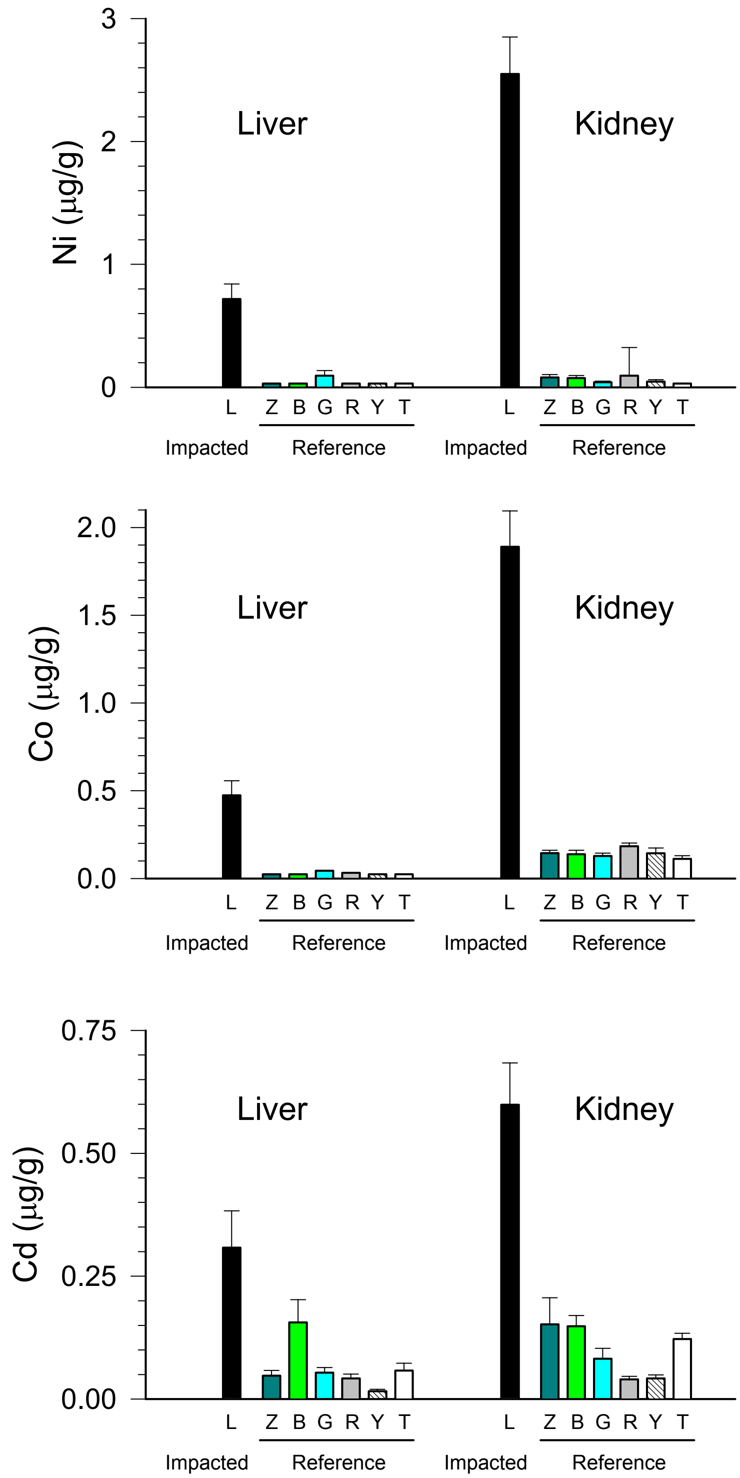

**Figure 5.** Linear regression analyses of Cu against Mo (upper panel) and of Hg against Se (lower panel) in kidneys of white suckers from Fox Lake

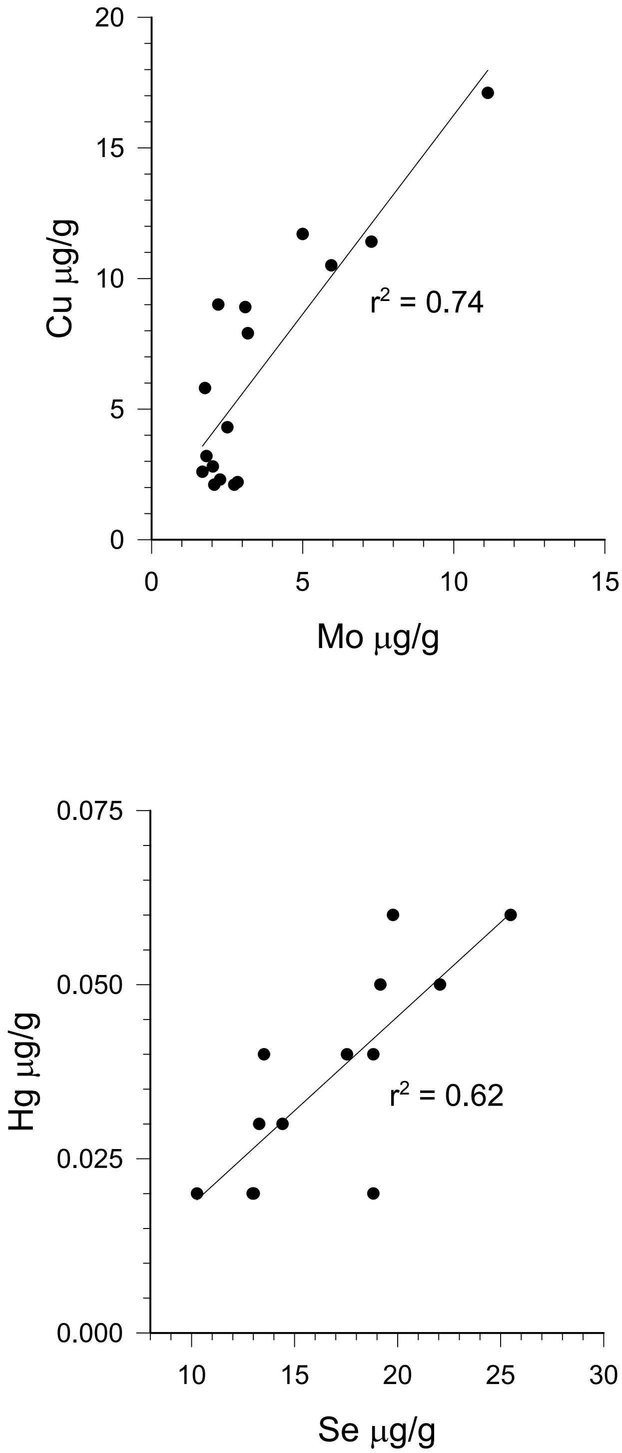

**Figure 6.** Concentrations (μg/g, wet wt.) of metallothionein in livers and kidneys of white suckers from Fox Lake and five reference lakes. Letters used to designate lakes are defined in Figure 3. Data are expressed as mean ± S.E.M. See Table 2 for n values.

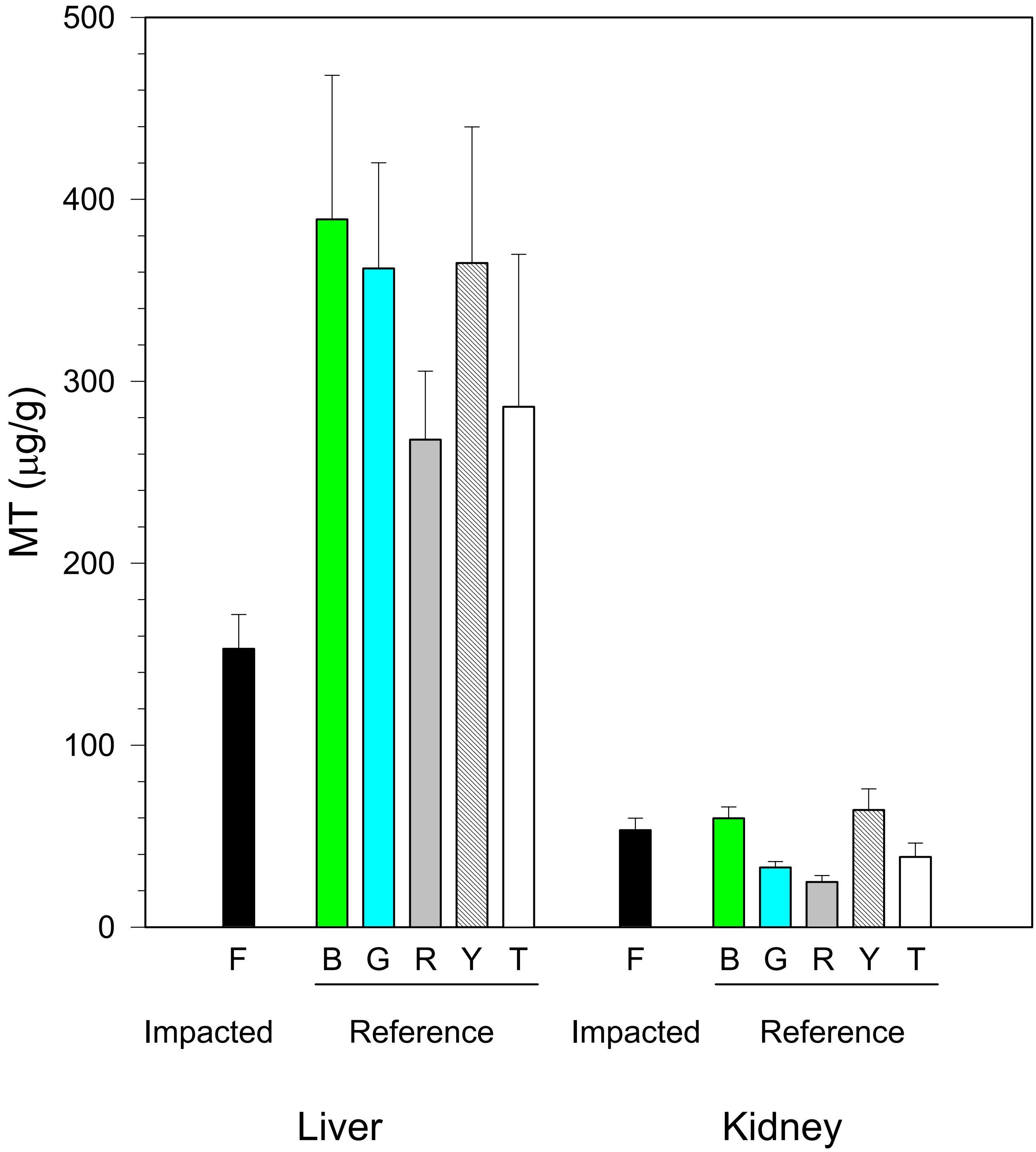

**Figure 7.** Linear regression analyses of metallothionein (MT) concentrations (μg/g, wet wt.) against Cu concentrations (μg/g, wet wt.) in livers of white suckers from Fox Lake (panel A), white suckers from the reference lakes (panel B), northern pike from reference lakes (panel C), and lake whitefish from Little McDonald Lake (panel D). In panel B reference lakes are denoted as (■) Bizarre Lake, (●) Boomerang Lake, (▲) Little Yalowega Lake, (○) Lower Read Lake, and (□) Toby Lake. In panel C reference lakes are denoted as in panel B, with the addition of (△) Zimmer Lake.

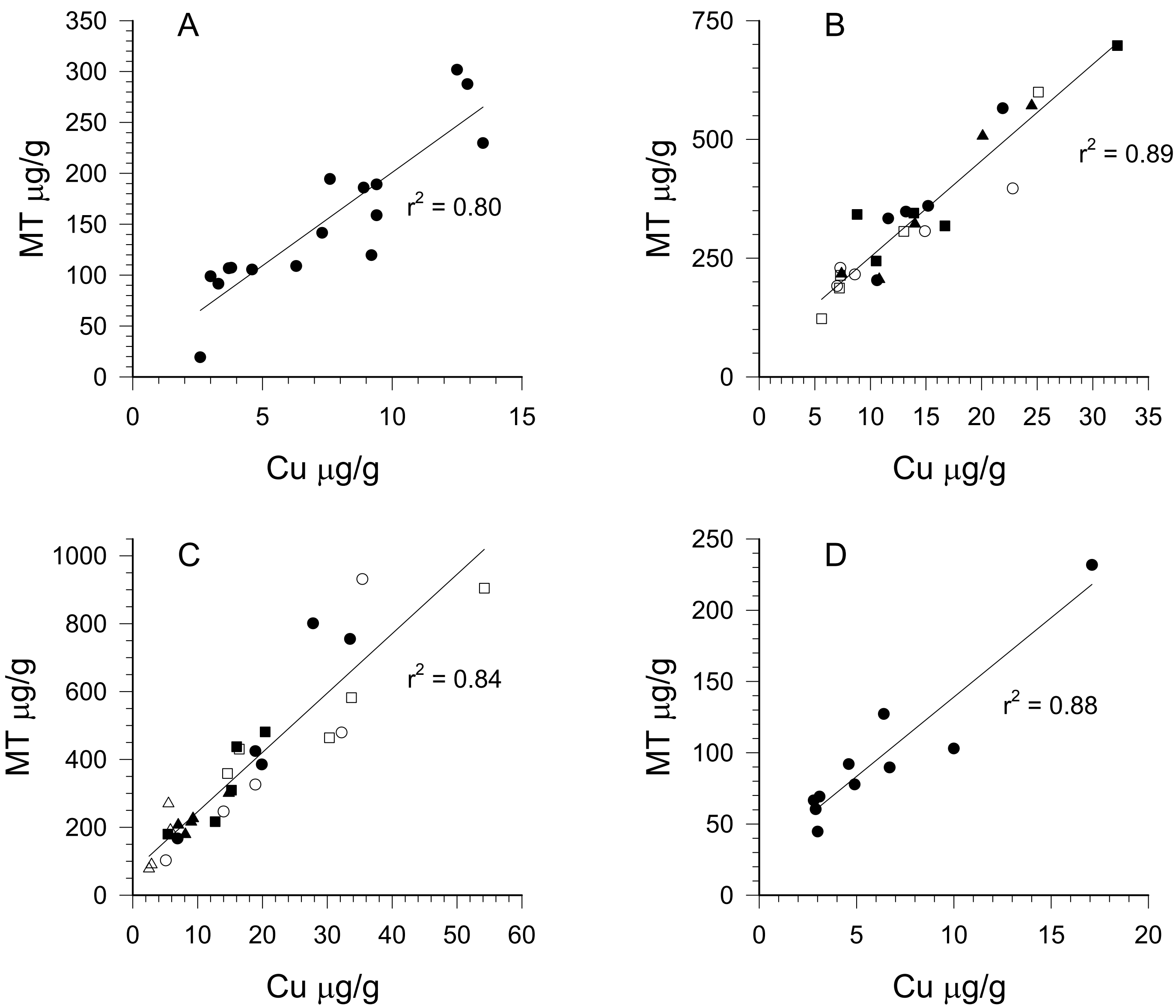

**Table 1.** Metal concentrations (μg/g, d.w.)in the surficial (top) section of sediment cores from Wolf Lake, Fox Lake and Little McDonald Lake; and the Enrichment Factor (EF) for each metal. Metal concentrations are expressed as mean ± S.E.M., n = 4. EFs are calculated as the ratio of metal concentration in the top 1 cm section to the concentration in the deepest section (20 cm).

| **Metals** (μg/g dry wt) | **Receiving Mill Effluent** | | **Receiving Dewatering Discharges** |
|---|---|---|---|
| | Wolf Lake | Fox Lake | Little McDonald Lake |
| Al | 6,870 ± 1,280<br>EF ≤ 1 | 7,820 ± 310<br>EF ≤ 1 | 21,700 ± 1,180<br>EF = 1.3 |
| As | 5,890 ± 1,120<br>EF = 259 | 3,830 ± 64<br>EF = 276 | 160 ± 23<br>EF = 3.9 |
| Cd | 0.27 ± 0.094<br>EF = 1.8 | 0.17 ± 0.08<br>EF ≤ 1 | 1.15 ± 0.06<br>EF ≤ 1 |
| Co | 6.0 ± 2.1<br>EF ≤ 1 | 12.4 ± 5.6<br>EF = 4.1 | 446 ± 119<br>EF = 29.5 |
| Cu | 18.4 ± 2.0<br>EF = 4.8 | 12.6 ± 1.0<br>EF = 2.7 | 21.8 ± 4.2<br>EF = 1.9 |
| Fe | 61,000 ± 2,900<br>EF = 13.6 | 100,700 ± 5,300<br>EF = 2.9 | 163,000 ± 42,000<br>EF = 1.2 |
| Hg | 0.071 ± 0.019<br>EF = 3.6 | 0.122 ± 0.028<br>EF = 2.7 | 0.094 ± 0.008<br>EF = 1.8 |
| Pb | 5.67 ± 1.9<br>EF = 3.6 | 10.1 ± 3.6<br>EF = 1.7 | 66.3 ± 17<br>EF = 3.4 |
| Mn | 10.7 ± 2.2<br>EF ≤ 1 | 28.1 ± 5.6<br>EF ≤ 1 | 14,300 ± 3,400<br>EF = 8.8 |
| Mo | 1,800 ± 280<br>EF = 720 | 3,980 ± 470<br>EF = 323 | 15.6 ± 5.4<br>EF = 3.1 |
| Ni | 129 ± 50<br>EF = 6.1 | 129 ± 14<br>EF = 5.2 | 946 ± 230<br>EF = 10.3 |
| Se | 22.8 ± 18<br>EF = 33.5 | 54.4 ± 4.6<br>EF = 25.8 | 0.91 ± 0.24<br>EF ≤ 1 |
| Zn | 27.5 ± 3.3<br>EF = 1.7 | 31.4 ± 3.6<br>EF ≤ 1 | 373 ± 56<br>EF = 2.5 |

**Table 2.** Fish meristics – 1994. Numbers (n) of fish sampled, fork lengths (cm), weights (g), ages (years), and sexes of white suckers, northern pike and lake whitefish. Data for fork lengths, weights and ages are expresses as mean ± S.E.M. Mean comparisons employed Dunnett's procedure one-tailed († ) and two-tailed (*), as indicated.

| Species | Lake | n | Fork Length (cm) | Weight (g) | Age (y) | Sex |
|---|---|---|---|---|---|---|
| **White Sucker** | Fox | 16 | 41.6 ± 0.8 | 1060 ± 57 | 11.1 ± 1.0 | 7 M : 9 F |
| | Bizarre | 5 | 47.3 ± 0.8 * | 1634 ± 118 * | 12.2 ± 2.4 | 3 M : 2 F |
| | Boomerang | 5 | 46.6 ± 1.4 * | 1426 ± 122 * | 11.4 ± 0.7 | 2 M : 3 F |
| | Lower Read | 5 | 44.8 ± 1.1 | 1324 ± 75 † | 14.4 ± 1.7 | 1 M : 4 F |
| | Little Yalowega | 5 | 41.6 ± 0.7 | 1048 ± 38 | 10.8 ± 1.0 | 5 F |
| | Toby | 5 | 42.6 ± 0.9 | 1126 ± 58 | 11.2 ± 0.7 | 2 M : 3 F |
| **Northern Pike** | Little McDonald | 11 | 55.2 ± 1.8 | 1174 ± 386 | 5.5 ± 0.39 | 7 M : 4 F |
| | Zimmer | 4 | 60.8 ± 4.4 | 1608 ± 397 | 5.8 ± 0.48 | 2 M : 2 F |
| | Bizarre | 5 | 55.9 ± 1.5 | 1084 ± 92 | 10.2 ± 0.86 * | 4 M : 1 F |
| | Boomerang | 5 | 70.9 ± 3.8 * | 2278 ± 336 † | 9.6 ± 0.51 * | 1 M : 4 F |
| | Lower Read | 5 | 73.4 ± 6.9 * | 2844 ± 787 * | 10.8 ± 1.6 * | 5 F |
| | Little Yalowega | 5 | 63.4 ± 1.2 | 1806 ± 87 | 6.8 ± 0.86 | 2 M : 3 F |
| | Toby | 5 | 57.7 ± 0.96 | 1304 ± 59 | 7.2 ± 0.6 | 3 M : 2 F |
| **Lake Whitefish** | Little McDonald | 10 | 39.2 ± 0.62 | 771 ± 39 | 8.9 ± 0.71 | 3 M : 7 F |
| | Zimmer | 4 | 36.3 ± 3.7 | 738 ± 73 | 13.0 ± 2.7 † | 3 M : 1 F |

† Dunnett's one-tailed $\alpha = 0.05$

* Dunnett's two-tailed $\alpha = 0.05$

**Table 3.** Regression equation parameters for significant ($p \leq 0.05$) relationships between fish age and metal concentrations in livers and kidneys, and between fish weight and metal concentrations in those tissues. Summary of regression analyses model **$[Mx] = \beta_0 + \beta_1(age) + \varepsilon_{ij}$**

**White Sucker**

| Location | Tissue | Mx | p-value | $r^2$ | $\beta_0$ | $\beta_1$ |
|---|---|---|---|---|---|---|
| impacted | liver | As | 0.0342 | 0.28 | 0.16 | 0.040 |
| impacted | liver | Cd | 0.0099 | 0.39 | -0.021 | 0.0046 |
| impacted | kidney | Cd | 0.0144 | 0.36 | -0.12 | 0.019 |
| impacted | liver | Cu | 0.0082 | 0.40 | 14 | -0.56 |
| reference | liver | Hg | 0.0001 | 0.50 | 0.0034 | 0.0049 |
| reference | kidney | Ni | 0.0435 | 0.17 | 0.011 | 0.0022 |
| reference | liver | Se | 0.0097 | 0.26 | 0.35 | 0.052 |
| impacted | liver | Zn | 0.0167 | 0.35 | 30 | -0.70 |

**Northern Pike**

| Location | Tissue | Mx | p-value | $r^2$ | $\beta_0$ | $\beta_1$ |
|---|---|---|---|---|---|---|
| reference | liver | Hg | 0.0159 | 0.21 | -0.014 | 0.019 |
| reference | kidney | Hg | 0.0002 | 0.42 | -0.040 | 0.022 |
| reference | kidney | Mn | 0.0148 | 0.21 | 1.1 | -0.049 |
| impacted | kidney | Ni | 0.0196 | 0.47 | -0.35 | 0.52 |
| reference | liver | Se | 0.0275 | 0.17 | 1.1 | 0.059 |

Lake Whitefish**:** $p > 0.05$ for all locations, tissues and metals

Summary of regression analyses model **$[Mx] = \beta_0 + \beta_1(weight) + \varepsilon_{ij}$**

**White Sucker**

| Location | Tissue | Mx | p-value | $r^2$ | $\beta_0$ | $\beta_1$ |
|---|---|---|---|---|---|---|
| reference | liver | Cd | 0.0152 | 0.23 | -0.022 | $8.28 \times 10^{-5}$ |
| impacted | liver | Cu | 0.0158 | 0.35 | 17 | -0.0095 |
| reference | kidney | Cu | 0.0231 | 0.20 | 3.2 | -0.0010 |
| reference | liver | Hg | 0.0082 | 0.27 | 0.0085 | $4.08 \times 10^{-5}$ |
| reference | kidney | Hg | 0.0046 | 0.30 | -0.0093 | $4.25 \times 10^{-5}$ |
| impacted | kidney | Mn | 0.0042 | 0.46 | 0.97 | $-1.93 \times 10^{-4}$ |

**Northern Pike**

| Location | Tissue | Mx | p-value | $r^2$ | $\beta_0$ | $\beta_1$ |
|---|---|---|---|---|---|---|
| reference | kidney | Cd | 0.0256 | 0.17 | 0.15 | $-2.73 \times 10^{-5}$ |
| reference | kidney | Hg | 0.0277 | 0.17 | 0.078 | $1.63 \times 10^{-5}$ |
| impacted | liver | Mn | 0.0327 | 0.41 | -0.21 | 0.0020 |

**Lake Whitefish**

| Location | Tissue | Mx | p-value | $r^2$ | $\beta_0$ | $\beta_1$ |
|---|---|---|---|---|---|---|
| reference | liver | Hg | 0.0020 | 1.0 | -0.72 | 0.0013 |
| impacted | liver | Mn | 0.0385 | 0.43 | -0.22 | 0.0033 |

**Table 4**. Concentrations (µg/g; wet wt.) of metals in livers and kidneys of white suckers: A summary of comparisons between Fox Lake and five reference lakes (RLs). Metal concentrations in tissues of white suckers from Fox Lake are expressed as mean ± S.E.M. See Table 2 for n values.

| Metal: | Tissue: | Conc. In Fox Lake White Suckers: | Comparisons of Fox Lake to RLs: |
|---|---|---|---|
| Mo | Liver | 2.1 ± 0.2 | 6.0 to 8.8 x higher than RLs |
| | Kidney | 3.6 ± 0.6 | 12.0 to 37.5 x higher than RLs |
| Se | Liver | 17.9 ± 0.8 | 12.2 to 23.9 x higher than RLs |
| | Kidney | 16.1 ± 1.2 | 19.2 to 32.3 x higher than RLs |
| Cu | Liver | 7.4 ± 0.9 | Lowest in Fox (45% to 64% of RLs) |
| | Kidney | 6.5 ± 1.1 | 2.3 to 5.1 x higher than RLs |
| As | Liver | 0.6 ± 0.1 | Fox = 15.3 x DL; all RLs were < DL |
| | Kidney | 47% < DL | All RLs were < DL |
| Ni | Liver | 0.11 ± 0.01 | Fox = 2 x DL; most RLs were < DL |
| | Kidney | 0.29 ± 0.04 | Fox = 5 x DL; most RLs were < DL |
| Zn | Liver | 21.8 ± 1.3 | Lowest in Fox (77% to 98% of RLs) |
| | Kidney | 17.6 ± 1.9 | Lowest in Fox (77% to 93% of RLs) |
| Cd | Liver | 44% < DL | Lowest in Fox (RLs were 2.2 to 8 x DL) |
| | Kidney | 31% < DL | Lowest in Fox (RLs were 1.1 to 9 x DL) |
| Hg | Liver | 0.028 ± 0.002 | Lowest in Fox (33% to 70% of RLs) |
| | Kidney | 0.037 ± 0.004 | Fox in middle of range |
| Mn | Liver | 1.2 ± 0.1 | Lowest in Fox (48% to 79% of RLs) |
| | Kidney | 0.3 ± 0.1 | Lowest in Fox (19% to 50% of RLs) |
| Fe | Liver | 259 ± 20 | Fox in middle of range |
| | Kidney | 99 ± 7 | 1.1 to 1.7 x higher than RLs |
| Co | Liver | < DL | All values were < DL |
| | Kidney | 63% < DL | Fox in middle of range |
| Pb | Liver | < DL | Most values were < DL |
| | Kidney | < DL | All values were < DL |

Reference Lakes = Bizarre, Boomerang, Little Yalowega, Lower Read, and Toby

Detection Limits (DL; µg/g, wet wt):

| | | |
|---|---|---|
| As = 0.04 | Ni = 0.06 | Cd = 0.02 |
| Co = 0.05 | Pb = 0.10 | |

**Table 5.** Concentrations (μg/g; wet wt.) of metals in livers and kidneys of northern pike: A summary of comparisons between Little McDonald (LMcD) Lake and six reference lakes (RLs). Metal concentrations in tissues of northern pike from Little McDonald Lake are expressed as mean ± S.E.M. See Table 2 for n values.

| Metal: | Tissue: | Conc. In LMcD Lake Northern Pike: | LMcD Lake Comparisons to RLs: |
|---|---|---|---|
| Mo | Liver | 0.30 ± 0.04 | 1.5 to 2.0 x higher than RLs |
| | Kidney | 0.17 ± 0.04 | 1.5 to 2.5 x higher than RLs |
| Se | Liver | 2.4 ± 0.1 | 1.2 to 1.8 x higher than RLs |
| | Kidney | 1.9 ± 0.3 | LMcD = second highest concentration |
| Cu | Liver | 10.9 ± 2.5 | LMcD in middle of range |
| | Kidney | 2.1 ± 0.2 | 1.6 to 2.8 x higher than RLs |
| As | Liver | 100% < DL | All values were < DL |
| | Kidney | 100% < DL | All values were < DL |
| Ni | Liver | 0.72 ± 0.12 | Most RL values were < DL |
| | Kidney | 2.6 ± 0.3 | 27 to 42.5 x higher than RLs |
| Zn | Liver | 60.0 ± 7.1 | LMcD = second highest concentration |
| | Kidney | 152 ± 16 | LMcD in middle of range |
| Cd | Liver | 0.31 ± 0.08 | 2 to 19.3 x higher than RLs |
| | Kidney | 0.60 ± 0.09 | 4 to 15 x higher than RLs |
| Hg | Liver | 0.27 ± 0.11 | 1.3 to 2.8 x higher than RLs |
| | Kidney | 0.34 ± 0.14 | 1.6 to 3.8 x higher than RLs |
| Mn | Liver | 2.2 ± 0.4 | 1.3 to 2.8 x higher than RLs |
| | Kidney | 1.4 ± 0.3 | 1.3 to 3.1 x higher than RLs |
| Fe | Liver | 317 ± 91 | LMcD in middle of range |
| | Kidney | 111 ± 12 | LMcD = second highest concentration |
| Co | Liver | 0.47 ± 0.08 | LMcD = 9.5 x DL; all RLs were < DL |
| | Kidney | 1.89 ± 0.20 | 13 to 17 x higher than RLs |
| Pb | Liver | < DL, (2 exceptions) | RLs were < DL with rare exceptions |
| | Kidney | < DL, (1 exception) | RLs were < DL with rare exceptions |

Reference Lakes = Bizarre, Boomerang, Little Yalowega, Lower Read, Toby and Zimmer

Detection Limits (DL; μg/g, wet wt.):

| | | |
|---|---|---|
| As = 0.04 | Ni = 0.06 | Cd = 0.02 |
| Co = 0.05 | Pb = 0.10 | |

**Table 6**. Concentrations (μg/g, wet wt.) of metals and metallothionein (MT) in livers and kidneys of lake whitefish from Little McDonald (LMcD) Lake and Zimmer Lake. Metal concentrations in tissues of lake whitefish from Little McDonald Lake are expressed as mean ± S.E.M.

| Analyte: | Tissue: | Conc. in LMcD Lake | Conc. in Zimmer L. | Ratio of LMcD :Zimmer L. |
|---|---|---|---|---|
| Ni | Liver | 0.565 ± 0.121 | < 0.06 | 9.4 |
| | Kidney | 4.66 ± 0.72 | 0.645 ± 0.192 | 7.2 |
| Mo | Liver | 0.195 ± 0.042 | 0.145 ± 0.018 | 1.34 |
| | Kidney | 0.125 ± 0.016 | 0.110 ± 0.015 | 1.14 |
| Co | Liver | 0.234 ± 0.053 | 0.068 ± 0.006 | 3.44 |
| | Kidney | 0.633 ± 0.108 | 0.562 ± 0.064 | 1.13 |
| Cd | Liver | 0.211 ± 0.045 | 0.272 ± 0.148 | 0.78 |
| | Kidney | 1.73 ± 0.41 | 1.96 ± 0.603 | 0.88 |
| Cu | Liver | 6.15 ± 1.41 | 7.62 ± 1.75 | 0.81 |
| | Kidney | 1.78 ± 0.13 | 1.25 ± 0.119 | 1.42 |
| Zn | Liver | 29.5 ± 2.8 | 29.0 ± 3.22 | 1.02 |
| | Kidney | 20.4 ± 1.7 | 24.6 ± 2.71 | 0.83 |
| Fe | Liver | 80.7 ± 19.6 | 393 ± 247 | 0.21 |
| | Kidney | 172 ± 32.2 | 452 ± 203 | 0.38 |
| Hg | Liver | 0.108 ± 0.017† | 0.233 ± 0.134 | 0.464 |
| | Kidney | 0.243 ± 0.055* | 0.615 ± 0.045 | 0.395 |
| Se | Liver | 2.72 ± 0.30† | 1.95 ± 0.364 | 1.395 |
| | Kidney | 2.15 ± 0.15† | 2.14 ± 0.375 | 1.005 |
| MT | Liver | 96.2 ± 16.8 | 182 ± 22.5 | 0.53 |
| | Kidney | 71.4 ± 16.6 | 70.1 ± 6.26 | 1.02 |

n = 10 from LMcD Lake, except where noted
n = 4 from Zimmer Lake
* n = 3
† n = 8

**Table 7.** Concentrations (μg/g, wet wt.) of metallothionein (MT) in livers and kidneys of northern pike from Little McDonald Lake and six reference lakes. Data are expressed as mean ± S.E.M.

| Lake | n | Liver MT | Kidney MT |
|---|---|---|---|
| Little McDonald | 11 | 264 ± 30 | 109 ± 37 |
| Zimmer | 4 | 158 ± 45 | 168 ± 37 |
| Bizarre | 5 | 325 ± 59 | 99 ± 10 |
| Boomerang | 5 | 506 ± 119 | 90 ± 26 |
| Lower Read | 5 | 417 ± 142 | 103 ± 23 |
| Little Yalowega | 5 | 226 ± 20 | 90 ± 12 |
| Toby | 5 | 548 ± 96 | 82 ± 25 |

**Table 8.** A summary of concentrations of water quality parameters as presented in Pyle et al. (2001) for a reference lake (David Lake) and for lakes impacted either by treated mill effluent (Fox Lake) or by treated mine dewatering discharges (Little McDonald Lake).

## Water Quality Parameters[1]

| Lake: | David[a] | Fox[b] | Little McDonald[c] |
|---|---|---|---|
| pH | | | |
| Median: | 6.1 | 5.1 | 6.7 |
| Range: | 5.8 – 6.3 | 4.7 – 5.4 | 6.3 – 6.8 |
| Hardness (as $CaCO_3$, mg/L) | 9.6 ± 1.6 | 687 ± 60 | 29.1 ± 8.1 |
| Alkalinity (as $CaCO_3$, mg/L) | 19.0 ± 1.4 | 22.2 ± 2.6 | 24.9 ± 2.0 |
| Dissolved $O_2$ (mg/L) | 7.3 ± 0.2 | 7.4 ± 0.1 | 8.7 ± 0.1 |
| Conductivity (μS/cm) | 23.0 ± 2.0 | 2112 ± 27 | 76.4 ± 2.6 |
| T.D.S.[2] (mg/L) | 11.6 ± 1.0 | 1057 ± 9 | 38.5 ± 1.4 |
| Carbon Dioxide (mg/L) | 5.3 ± 0.5 | 12.1 ± 2.0 | 4.6 ± 0.9 |
| Turbidity (F.T.U.) | 20.0 ± 4.0 | 33.1 ± 8.2 | 5.9 ± 1.0 |

[1]Published in Pyle et al., 2001. Ecotoxicol. Environ. Safety **48**: 202-214; With the exception of pH, data are expressed as mean ± S.E.M.
[2]T.D.S. = Total Dissolved Solids
[a] a reference lake
[b] a lake impacted by mill effluent
[c] a lake impacted by mine dewatering discharges

**Table 9.** Concentrations of metals (μg/L) in the waters of lakes described in Table 8.

## Concentrations[1] of Metals In Lake Waters[2]

| Lake: | David[a] | | Fox[b] | | Little McDonald[c] | |
|---|---|---|---|---|---|---|
| Metal | Total [Me] | % D[d] | Total [Me] | % D[d] | Total [Me] | % D[d] |
| As | 0.38 ± 0.04 | 79 | 43.66 ± 3.25 | 94 | 0.25 ± 0.03 | 68 |
| Cd | 0.09 ± 0.09 | 133 | 1.96 ± 0.13 | 97 | 0.05 ± 0.02 | 100 |
| Co | 0.05 ± 0.01 | 100 | 1.33 ± 0.05 | 98 | 2.28 ± 0.36 | 43 |
| Cu | 0.41 ± 0.14 | 139 | 1.29 ± 0.06 | 91 | 0.59 ± 0.05 | 134 |
| Fe | 356 ± 108 | 46 | 1238 ± 28 | 90 | 61 ± 7 | 43 |
| Pb | * | - | * | - | * | - |
| Hg | 0.01 ± 0.005 | 100 | 0.05 ± 0.02 | 40 | 0.02 ± 0.005 | 50 |
| Mn | 15.5 ± 3.3 | 74 | 67.6 ± 1.3 | 102 | 38.2 ± 4.5 | 46 |
| Mo | 0.39 ± 0.22 | 8 | 1397 ± 61 | 100 | 0.72 ± 0.39 | 1 |
| Ni | 0.54 ± 0.08 | 102 | 33.3 ± 1.7 | 96 | 116.0 ± 4.6 | 93 |
| Se | 0.46 ± 0.38 | 7 | 7.7 ± 0.7 | 77 | 0.73 ± 0.68 | 4 |
| Zn | 2.42 ± 1.31 | 110 | 1.47 ± 0.15 | 69 | 12. 03 ± 1.04 | 96 |

[1]Data (μg/L) are expressed as mean ± S.E.M.; n = 4
[2]Published in Pyle et al., 2001. Ecotoxicol Environ. Safety **48:** 202-214.
[a] a non-impacted reference lake
[b] a lake impacted by mill effluent
[c] a lake impacted by mine dewatering discharges
[d] % dissolved metal
* = no data were provided for Pb

**Table 10.** Concentrations (μg/g, wet wt.) of Cu, Zn, Cd and Hg in livers and kidneys of white suckers from natural populations. Data are expressed as mean ± S.E.M., except where noted.

| Metal: | Lake(s): | LIVERS | KIDNEYS | Reference: |
|---|---|---|---|---|
| Cu | Fox | 7.4 ± 0.9 (C) | 6.5 ± 1.1 (C) | This study |
| | Keg | 32.5 ± 4.5 (R) | 1.7 ± 0.1 (R) | Klaverkamp et al., 2002 |
| | 5 lakes near Flin Flon smelter | 20.2 ± 3.0 (C) | n.a. | Harrison & Klaverkamp, 1990 |
| | 3 lakes in N. Saskatchewan | 10.2 ± 4.4 (R) | n.a. | Harrison & Klaverkamp, 1990 |
| | Manitouwadge chain, Ontario | 10.0 ± 1.2 (R) | 2.8 ± 0.4 (R) | Munkittrick et al.,1991 |
| | " " " " " | 16.6 ± 1.2 (C) | 5.2 ± 0.8 (C) | " " " " " " |
| | Manitouwadge chain, Ontario | 10.2 ± 1.2 (R) | 1.6 ± 0.1 (R) | Miller et al., 1992 |
| | " " " " " | 19.6 ± 1.6 (C) | 6.2 ± 0.8 (C) | " " " " |
| | ELA lake 468 | 18.0 ± 0.8 (R) | 1.6 ± 0.1 (R) | Klaverkamp, 1999 unpub.observ. |
| | Moss (in Adirondack Park) | 5.8 ± 0.6 (C) | 7.0 ± 0.4 (C) | Stripp et al., 1990 |
| | Dart (in Adirondack Park) | 4.8 ± 0.5 (C) | 6.0 ± 0.8 (C) | " " " " |
| | 8 S.-Central Ontario lakes:<br>ranges of Rs to Cs: | 5.4 ± 0.6 to<br>16.6 ± 1.2 | 0.8 ± 0.1 to<br>2.0 ± 0.1 | Bendell-Young & Harvey, 1989 |
| | | | | |
| | | | | |
| Zn | Fox | 21.8 ± 1.3 (C) | 17.6 ± 1.9 (C) | This study |
| | Keg | 37.8 ± 2.2 (R) | 20.6 ± 1.5 (R) | Klaverkamp et al., 2002 |
| | 5 lakes near Flin Flon smelter | 43.0 ± 4.5 (C) | n.a. | Harrison & Klaverkamp, 1990 |
| | 3 lakes in N. Saskatchewan | 23.0 ± 4.0 (C) | n.a. | Harrison & Klaverkamp, 1990 |
| | Manitouwadge chain, Ontario | 22.4 ± 1.8 (R) | 19.4 ± 1.4 (R) | Munkittrick et al.,1991 |
| | " " " " " | 42.0 ± 2.8 (C) | 38.4 ± 3.4 (C) | " " " " " " |
| | Manitouwadge chain, Ontario | 24.0 ± 1.8 (R) | 22.4 ± 0.6 (R) | Miller et al., 1992 |
| | " " " " " | 55.0 ± 8.0 (C) | 53.8 ± 2.6 (C) | " " " " |
| | ELA lake 468 | 32.7 ± 2.1 (R) | 24.2 ± 1.3 (R) | Klaverkamp, 1999 unpub.observ. |
| | 8 S.-Central Ontario lakes: | | | Bendell-Young & Harvey, 1989 |
| | Ranges of Rs to Cs: | 14.0 ± 0.9 to | 18.4 ± 0.7 | |
| | | 30.6 ± 1.6 | 29.2 ± 1.6 | |
| | | | | |
| Cd | Fox | 44% < 0.02* | 31% < 0.02* | This study |
| | Keg | 0.18±0.03 (R) | 0.93±0.38 (R) | Klaverkamp et al., 2002 |
| | 5 lakes near Flin Flon smelter | 0.28±0.09 (C) | n.a. | Harrison & Klaverkamp, 1990 |
| | 3 lakes in N. Saskatchewan | 0.15±0.07 (R) | | Harrison & Klaverkamp, 1990 |
| | ELA lake 468 | 0.17±0.02 (R) | 1.10±0.23 (R) | Klaverkamp, 1999 unpub.observ. |
| | Moss (in Adirondack Park) | 0.04±0.00 (C) | 0.16±0.03 (C) | Stripp et al., 1990 |
| | Dart (in Adirondack Park) | 0.18±0.06 (C) | 0.30±0.04 (C) | " " " " |
| | | | | |
| | | | | |
| Hg | Fox | 0.04±0.00 (C) | 0.04±0.00 (C) | This study |
| | Keg | 0.04±0.01 (R) | 0.02±0.00 (R) | Klaverkamp et al., 2002 |
| | 5 lakes near Flin Flon smelter | 0.02±0.00 (C) | | Harrison & Klaverkamp, 1990 |
| | 3 lakes in N. Saskatchewan | 0.05±0.01 (R) | | Harrison & Klaverkamp, 1990 |
| | ELA lake 468 | 0.05±0.01 (R) | 0.10±0.05 (R) | Klaverkamp, 1990 unpub.observ. |

n.a. = not analyzed *Cd detection limit of 0.02 μg/g
(R) = fish collected from a reference lake
(C) = fish collected from a contaminated lake

**Table 11.** Concentrations (μg/g, wet wt.) of Cu, Zn, Cd and Hg in livers and kidneys of northern pike from natural populations. Data are expressed as mean ± S.E.M., except where noted.

| Metal: | Lake(s): | LIVERS | KIDNEYS | Reference: |
|---|---|---|---|---|
| Cu | Little McDonald | 10.9 ± 2.5 (C) | 2.1 ± 0.2 (C) | This study |
| | Yellowknife Bay, Great Slave | 1.69 ± 0.20 (R) | 1.0 ± 0.1 (R) | Jackson et al., 1996 |
| | Keg | 11.6 ± 1.6 (R) | 1.0 ± 0.1 (R) | Klaverkamp et al., 2002 |
| | 5 lakes near Flin Flon smelter | 16.9 ± 3.6 (C) | n.a. | Harrison & Klaverkamp, 1990 |
| | 4 lakes in N. Saskatchewan | 19.5 ± 4.4 (R) | n.a. | “ “ “ “ “ “ “ |
| | Lake near Russian smelter | 17.4[a] (C) | 1.0[a] (C) | Kashulin & Reshetnikov, 1995 |
| | Ref. lake for Russian smelter | 2.3[a] (R) | 1.2[a] (R) | “ “ “ “ “ “ “ |
| | | | | |
| | | | | |
| Zn | Little McDonald | 60.0 ± 7.1 (C) | 152 ± 16 (C) | This study |
| | Yellowknife Bay, Great Slave | 21.6 ± 1.5 (R) | 48.1 ± 25.1 (R) | Jackson et al., 1996 |
| | Keg | 44.6 ± 6.6 (R) | 129.7± 14.6 (R) | Klaverkamp et al., 2002 |
| | 5 lakes near Flin Flon smelter | 62.0 ± 10.3 (C) | n.a. | Harrison & Klaverkamp, 1990 |
| | 4 lakes in N. Saskatchewan | 60.0 ± 12.5 (R) | n.a. | “ “ “ “ “ “ “ |
| | Lake near Russian smelter | 41.8[a] (C) | 92.3[a] (C) | Kashulin & Reshetnikov, 1995 |
| | Ref. lake for Russian smelter | 40.3[a] (R) | 95.6[a] (R) | “ “ “ “ “ “ “ |
| | | | | |
| | | | | |
| Cd | Little McDonald | 0.31 ± 0.08 (C) | 0.60 ± 0.09 (C) | This study |
| | Yellowknife Bay, Great Slave | 0.04 ± 0.01 (R) | 0.29 ± 0.05 (R) | Jackson et al., 1996 |
| | Keg | 0.03 ± 0.01 (R) | 0.12 ± 0.01 (R) | Klaverkamp et al., 2002 |
| | 5 lakes near Flin Flon smelter | 0.55 ± 0.17 (C) | n.a. | Harrison & Klaverkamp, 1990 |
| | 4 lakes in N. Saskatchewan | 0.30 ± 0.13 (R) | n.a. | “ “ “ “ “ “ “ |
| | Rivers in eastern England | | | Barak & Mason, 1990 |
| | range: | 0.03±0.01(R) to | n.a. | |
| | | 0.09 ± 0.04 (R) | n.a. | |
| | | | | |
| | | | | |
| Hg | Little McDonald | 0.27 ± 0.11 (C) | 0.34 ± 0.14 (C) | This study |
| | Yellowknife Bay, Great Slave | 0.05 ± 0.01 (R) | 0.15 ± 0.04 (R) | Jackson et al., 1996 |
| | Keg | 0.07 ± 0.01 (R) | 0.07 ± 0.02 (R) | Klaverkamp et al., 2002 |
| | 5 lakes near Flin Flon smelter | 0.05 ± 0.01 (C) | n.a. | Harrison & Klaverkamp, 1990 |
| | 4 lakes in N. Saskatchewan | 0.57 ± 0.17 (R) | n.a. | “ “ “ “ “ “ “ |
| | Rivers in eastern England | | | Barak & Mason, 1990 |
| | range: | 0.05±0.01(R) to | n.a. | |
| | | 0.13 ± 0.05(R) | n.a. | |

n.a. = not analyzed
[a] = only mean values were reported
(R) = fish collected from a reference site
(C) = fish collected from a contaminated site

**Table 12.** Concentrations (μg/g, wet wt.) of Cu, Zn, Cd and Hg in livers and kidneys of lake whitefish from natural populations. Data are expressed as mean ± S.E.M., except where noted.

| Metal: | Lake(s): | LIVERS | KIDNEYS | Reference: |
|---|---|---|---|---|
| Cu | Little McDonald | 6.15 ± 1.41 (C) | 1.78 ± 0.13 (C) | This study |
| | Yellowknife Bay, Great Slave | 6.68 ± 1.18 (R) | 0.80 ± 0.07 (R) | Jackson et al., 1996 |
| | Lake near Russian smelter | 9.16[a](C) | 5.44[a] (C) | Kashulin & Reshetnikov, 1995 |
| | Ref. lake for Russian smelter | 11.12[a] (R) | 7.6[a] (R) | " " " " " " " |
| | | | | |
| | | | | |
| Zn | Little McDonald | 29.5 ± 2.8 (C) | 20.4 ± 1.7 (C) | This study |
| | Yellowknife Bay, Great Slave | 27.6 ± 1.0 (R) | 22.6 ± 1.3 (R) | Jackson et al., 1996 |
| | Lake near Russian smelter | 24.4[a](C) | 58.8[a] (C) | Kashulin & Reshetnikov, 1995 |
| | Ref. lake for Russian smelter | 52.8[a] (R) | 64.4[a] (R) | " " " " " " " |
| | | | | |
| | | | | |
| Cd | Little McDonald | 0.21 ± 0.05 (C) | 1.73 ± 0.41 (C) | This study |
| | Yellowknife Bay, Great Slave | 0.11 ± 0.01 (R) | 0.60 ± 0.05 (R) | Jackson et al., 1996 |
| | | | | |
| | | | | |
| Hg | Little McDonald | 0.11 ± 0.02 (C) | 0.24 ± 0.06 (C) | This study |
| | Yellowknife Bay, Great Slave | 0.08 ± 0.01 (R) | 0.08 ± 0.01 (R) | Jackson et al., 1996 |

n.a. = not analyzed
(R) = fish collected from a reference site
(C) = fish collected from a contaminated site

[a] = only mean values were reported

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## APPENDIX

This appendix presents additional results on concentrations of Cr, U and V, in sediments from lakes impacted at the Key Lake mine site; and on concentrations of metals in Wollaston Lake sediments affected by effluent discharges from the Rabbit Lake mine site in northeastern Saskatchewan. The additional analyses were conducted on sediments from lakes at the Key Lake, but not on the fish tissues due to lack of sufficient liver or kidney tissue. Fish were not collected from Wollaston Lake, so metal analyses were limited to sediments from impacted sites in that lake. Sections of one core from each of Fox and Little McDonald Lakes were analyzed for $^{137}Cs$, $^{210}Pb$ and $^{226}Ra$. These data are available through the Freshwater Institute, Radionuclide Laboratory (Paul Wilkinson).

### Cr, U AND V AT KEY LAKE MINE SITE

Sediment cores were obtained from sites described in the main report in Wolf and Fox lakes and in the northwest bay of Little McDonald Lake in August 1994 by SCUBA divers. Cores were sectioned, transported, stored and analyzed in the Environmental Chemistry Laboratory at the Freshwater Institute in the manner described in the main body of this report.

Means of the metal concentrations in each section from the same depth were calculated from the cores sampled from each sampling site. Data ($\mu$g/g, dry wt.) were plotted (not shown) as described in the main body of this report as depth profiles of mean metal concentrations in the sediments, along with the associated standard errors. Enrichment factors (EF) were calculated, as described in the main body of the report, as the ratio of concentration in the top 1 cm section to the concentration in the deepest section. Table A-1 shows mean concentrations of Cr, U and V and their associated EFs for the impacted lakes at the Key Lake mine site. Sediment EFs were greatest for Cr and V in Wolf Lake, while U enrichment was greatest in surficial sediments in Little McDonald Lake.

### METALS AT RABBIT LAKE MINE SITE

The Rabbit Lake mine (58° 15' N; 103° 40' W) located 350 km north of La Ronge on the southwest shore of Wollaston Lake, Saskatchewan, commenced milling in June, 1975. From 1993 to 1996, the Rabbit Lake mine was licensed to produce 5.4 million kg of uranium annually. Mill tailings are disposed of in the mined-out Rabbit Lake open pit which was converted to a "pervious surround" tailings repository in which groundwater flows are directed to the periphery of the pit so as to minimize contaminant movement out of the tailings.

Treated mill and minewater effluents are discharged to Horseshoe Creek (drainage area of 27.5 $km^2$), which flows into Hidden Bay on Wollaston Lake (Fig. A-1). Horseshoe Creek is estimated to have a mean natural annual flow rate of 0.2 $m^3/s$ at its entry to Hidden Bay with a range from 0.06 $m^3/s$ to 0.6 $m^3/s$ between low flows and the ten year flood level. The average discharge of treated effluent to Horseshoe Creek, during the period of 1985 to 1994, was 0.098 $m^3/s$ (TAEM, 1996). Hidden Bay has a mean depth of 6.8 m, a maximum depth of 19m, a surface area of 2177 ha, a volume of 120 x $10^6 m^3$, and a shore length of

60 km (TAEM, 1996). The flushing rate of Hidden Bay is estimated to be 1.7 times/year with a mean residence time of 215 days (TAEM, 1996).

A small amount of seepage (estimated to be 0.8 L/s) from the effluent treatment plant reports to Parks Lake (Saskmont Engineering, 1982). The drainage area of Parks Lake is estimated to be 14 $km^2$ which would yield an estimated discharge of 0.09 $m^3/s$. The volume of Parks Lake is estimated to be $5.18 \times 10^6 m^3$ with a residence time of 664 days and a flushing rate of 0.55 times/year.

Sediment cores were obtained in August 1994 by SCUBA divers at two sites in Hidden Bay of Wollaston lake. These two sites were where Horseshoe Creek enters Hidden Bay, and at a site approximately 4 km east where Parks Creek drains into Hidden Bay. Water depths at these sites were 1.5 m (Horseshoe Creek inflow) and 5.0 m (Parks Creek inflow). Four cores were obtained from the Horseshoe Creek site and two cores were obtained at the Parks Creek site. Cores were sectioned from top to bottom on site into 1-cm sections to a depth of 10 cm, and into 2-cm sections to a depth of 20 cm or deeper immediately following collection. Individual sections were placed in labeled plastic (Whirlpak®) bags, air was excluded, and sections were immediately frozen on dry ice and transported to the Freshwater Institute in Winnipeg, MB. Sediment samples were prepared by lyophilization at $-50^o$ C for three to four days using a Labconco Lyph-lock 12 freeze-dryer.

At the Rabbit Lake site, Horseshoe Creek courses for 7 km through wetlands, which allow for considerable contaminant deposition within this part of the watershed before discharging into Hidden Bay of Wollaston Lake. Enrichment Factors at the mouth of Horseshoe Creek (Table A-2) were generally less than those observed in lakes at the Key Lake site where the flow path provides little possibility for deposition before discharge into receptor lakes. Enrichment factors of 60, 12.8, 8.6, 8.6 and 3.7 were observed at this site for Mo, U, As, Se and Cu, respectively. At the Parks Creek reference site, enrichment factors for these metals were 4.3, 1, 2, 2.6 and 1, respectively.

In impacted lakes at the Key Lake mine site and in Hidden Bay of Wollaston Lake, the following metals were analyzed in sections of sediment cores: Al, As, Cd, Co, Cu, Fe, Mn, Mo, Pb, Se and Zn. Comparing mean concentrations in the surficial sections of these cores (Table 1 and A-2) reveals both differences and similarities between sites and lakes. Aluminum concentrations in surficial sediments from Wollaston Lake were higher than comparable concentrations in affected lakes at the Key Lake mine site. Arsenic concentrations were from 1 to 3 orders of magnitude higher in the surficial sediments from the three lakes at the Key Lake mine site. Cadmium concentrations were about the same in Wolf, Fox and Wollaston Lakes. Relative to these Cd concentrations, Little McDonald surficial sediments contained 3.2 to 6.8 times higher Cd Concentrations. Cobalt concentrations were similar between the three lakes at the Key Lake mine site and the two sites in Wollaston Lake. Similar to Cd, Co concentrations were higher in surficial sediments from Little McDonald Lake. Copper concentrations were within the same order of magnitude between lakes and sites. Iron concentrations were from 1.6 to about

8 times higher in sediments from lakes at the Key Lake mine site. Manganese concentrations in surficial sediments at the Parks Creek inflow site were about 19 times higher than those measured at the Horseshoe Creek inflow site. Comparable concentrations in Little McDonald Lake sediments were from approximately 510 to 1,340 times higher than those observed in Wolf and Fox Lakes. The descending order of Mo concentrations was Fox Lake > Wolf Lake > Horseshoe Creek inflow > Parks Creek inflow ≅ Little McDonald Lake. The highest Pb concentrations were observed in surficial sediments from Little McDonald Lake and Parks Creek inflow. The descending order of Se concentrations was the same as for Mo. The descending order of Zn concentrations in surficial sediments was Little McDonald Lake > Parks Creek inflow > Fox Lake ≅ Wolf Lake ≅ Horseshoe Creek inflow.

Future research efforts at impacted and reference sites in Wollaston Lake should include expanding the analyses to include mercury and radionuclides, determining the bioavailability of metals for fishes, and measuring whether the fishes are responding to those contaminants. Because Wollaston Lake is huge (2,681 $km^2$ in surface area), emphasis should be placed on local, small-bodied fishes that are generally limited to smaller habitat areas than mobile, large-bodied fish species. As recommended in the main body of this report, measuring responses to these contaminants should include metallothionein, indicators of lipid peroxidation, and histopathological analyses of vital organs.

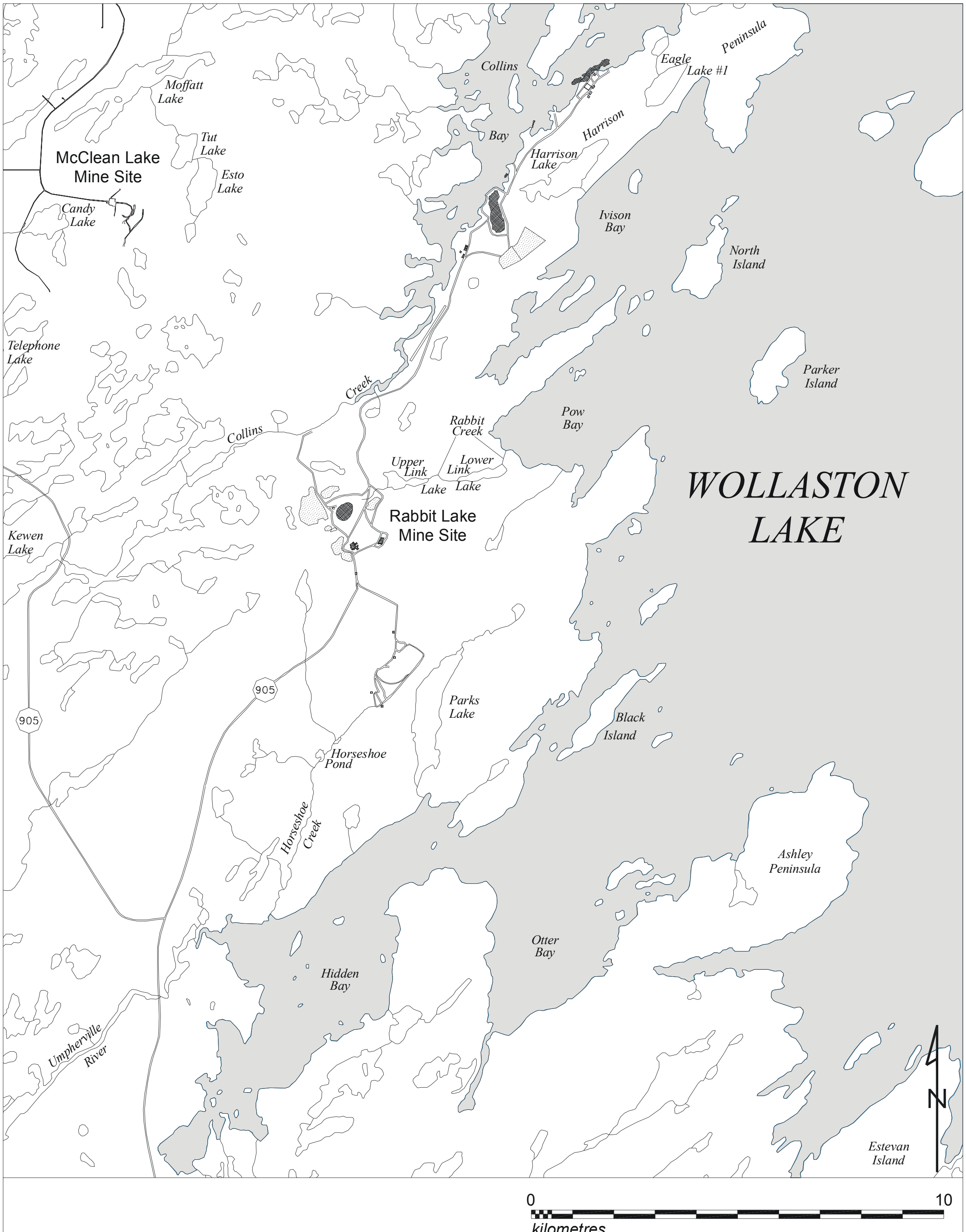


Figure A1
Rabbit Lake Project Area.

**Table A-1.** Concentrations of Cr, U and V (μg/g d.w.) in the surficial section of sediment cores from Wolf Lake, Fox Lake and Little McDonald Lake; and the Enrichment Factor (EF) for each metal. Metal concentrations are expressed as mean ± S.E.M., n = 4. EFs are calculated as the ratio of metal concentration in the top 1 cm section to the concentration in the deepest section (20 cm).

| **Metals** | **Receiving: Mill Effluent** | | **Dewatering Discharges** |
|---|---|---|---|
| (μg/g dry wt) | Wolf Lake | Fox Lake | Little McDonald Lake |
| Cr | 130.5 ± 16.1<br>EF = 8.0 | 94.0 ± 3.6<br>EF = 4.7 | 21.8 ± 3.0<br>EF ≤ 1 |
| U | 58.7 ± 22.9<br>EF = 1.4 | 93.0 ± 21.7<br>EF = 2.2 | 1,067 ± 255<br>EF = 4.8 |
| V | 88.8 ± 7.5<br>EF = 6.1 | 59.1 ± 4.4<br>EF = 2.6 | 34.1 ± 4.0<br>EF = 1.1 |

**Table A-2.** Metal concentrations (µg/g d.w.) in the surficial section of sediment cores from Hidden Bay in Wollaston Lake. Metal concentrations are expressed as mean ±S.E.M., with n=4 at the Horseshoe Creek inflow site and n=2 at the Parks Creek inflow site. EFs are calculated as the ratio of metal concentration in the top 1 cm section to the concentration in the deepest section (20 cm).

| Metals (µg/g dry wt) | Horseshoe Creek Inflow | Parks Creek Inflow |
|---|---|---|
| Al | 39,250 ± 3,400<br>EF ≤ 1 | 40,700 ± 2,300<br>EF ≤ 1 |
| As | 17.2 ± 1.0<br>EF = 8.6 | 4.0 ± 0.8<br>EF = 2.0 |
| Ba | 714 ± 140<br>EF ≤ 1 | 556 ± 98<br>EF ≤ 1 |
| Cd | 0.30 ± 0.05<br>EF = 1.3 | 0.36 ± 0.01<br>EF = 3.3 |
| Co | < 6.0 | 12 ± 3.0<br>EF = 1.3 |
| Cr | 26.5 ± 3.0<br>EF ≤ 1 | 26 ± 2.0<br>EF ≤ 1 |
| Cu | 22.1 ± 12.3<br>EF = 3.7 | 15.6 ± 5.0<br>EF = 2.6 |
| Fe | 20,750 ± 1,440<br>EF = 2.2 | 37,000 ± 1,000<br>EF = 2.1 |
| Mn | 157 ± 6.0<br>EF ≤ 1 | 2,930 ± 660<br>EF = 5.9 |
| Mo | 529 ± 55<br>EF = 60 | 21.5 ± 3.5<br>EF = 4.3 |
| Pb | 14.2 ± 4.2<br>EF ≤ 1 | 57.6 ± 12.5<br>EF ≤ 1 |
| Se | 3.6 ± 1.2<br>EF = 8.6 | < 1.0 |
| U | 639 ± 85<br>EF = 12.8 | < 50 |
| V | 21.8 ± 2.5<br>EF = 1.1 | 28 ± 1.0<br>EF = 1.1 |
| Zn | 23.5 ± 6.0<br>EF ≤ 1 | 67.8 ± 3.8<br>EF = 1.9 |

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