



Defining fish community structure in Lake Winnipeg using stable isotopes ($\delta^{13}\text{C}$, $\delta^{15}\text{N}$, $\delta^{34}\text{S}$): Implications for monitoring ecological responses and trophodynamics of mercury & other trace elements



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HIGHLIGHTS

- Anthropogenic eutrophication & non-native species affect Lake Winnipeg's ecosystem.
- We measured stable isotopes and trace elements in 15 native fish species.
- There was more evidence for growth dilution than biomagnification for most elements.
- The trophic structures of the north and south basins were different.
- These results will help determine the effects of recent arrival of zebra mussels.

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ABSTRACT

The ecological integrity of freshwater lakes is influenced by atmospheric and riverine deposition of contaminants, shoreline development, eutrophication, and the introduction of non-native species. Changes to the trophic structure of Lake Winnipeg, Canada, and consequently, the concentrations of contaminants and trace elements measured in tissues of native fishes, are likely attributed to agricultural runoff from the 977,800 km² watershed and the arrival of non-native zooplankters and fishes. We measured $\delta^{13}\text{C}$, $\delta^{15}\text{N}$, and $\delta^{34}\text{S}$ along with concentrations of 15 trace elements in 17 native fishes from the north and south basins of Lake Winnipeg in 2009 and 2010. After adjusting for differences in isotopic baseline values between the two basins, fishes in the south basin had consistently higher $\delta^{13}\text{C}$ and $\delta^{34}\text{S}$, and lower $\delta^{15}\text{N}$. We found little evidence of biomagnification of trace elements at the community level, but walleye (*Sander vitreus*) and freshwater drum (*Aplodinotus grunniens*) had higher mercury and selenium concentrations with increased trophic position, coincident with increased piscivory. There was evidence of growth dilution of cobalt, copper, manganese, molybdenum, thallium, and vanadium, and bioaccumulation of mercury, which could be explained by increases in algal (and consequently, lake and fish) productivity. We conclude that the north and south basins of Lake Winnipeg represent very different communities with different trophic structures and trace element concentrations.

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1. Introduction

Aquatic ecosystems throughout the world are affected by agricultural, urban, and industrial developments (Schindler, 1987; Sharpley et al., 1994; Zeng and Arnold, 2014). Associated changes to the relative abundance of nutrients entering lacustrine environments can affect the

numbers and nutritional quality of organisms at the base of the food web, thereby affecting the overall condition of upper-level consumers (Hebert et al., 2006). Increased atmospheric deposition and riverine loading of contaminants to lakes, estuaries and oceans have also raised concerns about the integrity of aquatic communities (Brumbaugh et al., 2005; Campbell et al., 2005; Ikemoto et al., 2008). In many instances, altered trophic interactions among aquatic biota have been concomitant with pollution of their habitats (Gewurtz et al., 2006; Ikemoto et al., 2008). Changes in food web structure and trophodynamics may modulate contaminant biomagnification and the rates at which toxic compounds accumulate in biota (Gustin et al., 2005; Jardine et al., 2006; Lavoie et al., 2013). For example, increases in lake productivity following nutrient enrichment and shifts in species diversity or abundance may

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significantly alter the availability of contaminants as well as the trophic interactions which facilitate their dietary transport (Stewart et al., 2003). Permanent changes in fish community structure may also occur in response to the introduction of non-native organisms or over-exploitation, and potentially depletion, of predatory fish populations (Bocking, 1997; Munawar et al., 2005) (Cucherousset et al., 2012). Such gains or losses of trophic linkages may ultimately decrease or increase the rates at which mercury (Hg) and other toxic compounds biomagnify through aquatic food webs (Jardine et al., 2006).

Lake Winnipeg, Canada (53° 17'N, 97° 58'W; Fig. 1) is the 8th-largest freshwater lake in the world (24,514 km²), and its food web is subject to numerous environmental and anthropogenic stressors (Environment Canada and Manitoba Water Stewardship, 2011). Floodwaters and runoff from livestock operations, cropland, urban centers and industrial areas within the 977,800 km² watershed contributed excess nutrients to the lake, resulting in eutrophication (Environment Canada and Manitoba Water Stewardship, 2011; Pip, 2006). Although much of the lake's nutrient load enters the southern basin via the Red River, the environment of the north basin supports conditions favorable for algal bloom formation (Pip, 2006). Other perturbations in the food web include changes in the relative abundance of plankton species and establishment of non-native zooplankters, as well as the introduction of zebra mussels (*Dreissena polymorpha*) (Manitoba Conservation and Water Stewardship, 2013; Stewart et al., 2003; Suchy et al., 2010) and populations of non-native fishes, such as common carp (*Cyprinus carpio*), white bass (*Morone chrysops*) and rainbow smelt (*Osmerus mordax*) (Stewart and Watkinson, 2004).

There has been considerable work over the past 10–15 years on organic pollutants in Lake Winnipeg (Gewurtz et al., 2006; Law et al., 2006; Stewart et al., 2003; Tomy et al., 2007). Few studies of Hg trophodynamics exist for Lake Winnipeg, despite the fact that elevated Hg concentrations in northern pike (*Esox lucius*), yellow perch (*Perca flavescens*), sauger (*Sander canadensis*), walleye (*Sander vitreus*) and freshwater drum (*Aplodinotus grunniens*) led to a season-long commercial fishing ban in the 1970s (Environment Canada and Manitoba Water Stewardship, 2011). Many trophic interactions among Lake Winnipeg fishes, and their effects on metal concentrations in fish tissues, have not been fully evaluated; this emphasizes the ongoing need to monitor the health of Lake Winnipeg (Environment Canada and Manitoba Water Stewardship, 2011; Hobson et al., 2012).

Trophic interactions among aquatic organisms are often inferred from nitrogen ($\delta^{15}\text{N}$) and carbon ($\delta^{13}\text{C}$) stable isotope analyses (Hebert et al., 2006; Hobson et al., 2012). With each trophic step, the $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ values of consumer tissues increase by approximately 2 to 5‰ and 1‰, respectively (Fry, 1988; Kelly, 2000). Since $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ values are reliable measures of trophic position and food source, respectively, they can help to identify contaminant sources, pathways, and rates of biomagnification (Campbell et al., 2005; Ikemoto et al., 2008; Jardine et al., 2006; Lavoie et al., 2013). Consumers that obtain energy and contaminants from pelagic versus benthic food sources can be separated based on tissue $\delta^{13}\text{C}$ values (Burgess and Hobson, 2006). Stable sulfur isotope ($\delta^{34}\text{S}$) measurements of tissues are also used to track food sources, namely sulfate derived from the water column or detritus and sediment-based foods (Croisetière et al., 2009; Wayland and Hobson, 2001).

Previous isotopic investigations of Lake Winnipeg's food web structure found considerable differences in the isotopic baseline of the two basins (Hobson et al., 2012), notable contributions of benthic algae to higher-trophic predators (Hecky and Hesslein, 1995), and patterns of bioaccumulation and biomagnification of contaminants (Law et al., 2006; Ofukany et al., 2012).

Our objective was to build upon this previous work, and to further evaluate the trophodynamics of Hg and other trace elements. We hypothesized that some elements, such as Hg, would be strongly correlated with $\delta^{15}\text{N}$, and therefore trophic position, while others (e.g., cadmium) might be related to a species' characteristically pelagic, benthic or detrital feeding habits ($\delta^{13}\text{C}$, $\delta^{34}\text{S}$).

2. Methods

Fish were collected from Lake Winnipeg during the spring (April–May), summer (June–August) and fall (September–November) of 2009 and 2010. Fish were also collected from the south basin during the winter of 2009–2010. A total of 16 fish species with variable fork lengths (FL; mm) were obtained from beam trawls and commercial gillnets (Supplementary material, Table S1). Beam trawls were towed alongside the *MV Namao* research vessel for 30-minute intervals and at a speed of 3.9 km/h (Lumb et al., 2012). Each station in Fig. 1 (closed circles) represents the ship's position at the end of each trawl, and was trawled three times (spring, summer and fall) per year. Commercial gillnets were used during winter of 2009–2010 (south basin only; Fig. 1; closed squares) and the ice-free season (spring, summer and fall; Fig. 1; closed triangles) of 2010 only. Gillnets were set in the evening and the catch was removed within 8 h (Johnston et al., 2012). Small specimens were frozen whole; however, one or both filets were removed from larger specimens, and placed in polyethylene bags (Bodaly et al., 2007; Hobson et al., 2012). All samples were held at $-20\text{ }^{\circ}\text{C}$ until they could be processed at the National Hydrology Research Centre in Saskatoon, Canada.

Samples were partially thawed and white muscle was carefully excised from each of the small, whole specimens (Fry, 1988). A stainless steel scalpel was used to make clean cuts into the dorsal muscle and extract 5 to 10 g of material (Pruell et al., 2003). A subsample of muscle from each specimen was reserved for isotopic assays. Portions of muscle from randomly-selected specimens were also set aside for elemental and Hg analyses.

2.1. Stable isotope analyses

Muscle samples were oven dried at 60 °C and homogenized (Vander Zanden et al., 2005). Nitrogen ($\delta^{15}\text{N}$), carbon ($\delta^{13}\text{C}$) and sulfur ($\delta^{34}\text{S}$) isotope values were quantified for separate aliquots of fish tissue (see below). Tissues used in $\delta^{13}\text{C}$ analyses were lipid extracted with a 2:1 chloroform:methanol solution (Bligh and Dyer, 1959). Lipid extraction can also remove muscle proteins, thereby confounding the results of $\delta^{15}\text{N}$ assays (Sotiropoulos et al., 2004), and since the effects of extraction on $\delta^{34}\text{S}$ have not been characterized, tissues used for $\delta^{15}\text{N}$ and $\delta^{34}\text{S}$ analyses were not lipid extracted (Hobson et al., 2012).

Subsamples (1.00 ± 0.01 mg) from each of untreated and lipid-extracted muscle were weighed into tin capsules for $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ isotope analyses, respectively (Vander Zanden et al., 2005), and 10.00 ± 0.10 mg of untreated muscle was used for $\delta^{34}\text{S}$ measurements. All stable isotope values were analyzed using continuous-flow isotope-ratio mass spectrometry (Wayland and Hobson, 2001). Laboratory measurement standards with assigned values that were carefully calibrated to international standards; BWB ($\delta^{15}\text{N} = +14.4\text{‰}$, $\delta^{13}\text{C} = -18.5\text{‰}$, $\delta^{34}\text{S} = +17.5\text{‰}$), PUGEL ($\delta^{15}\text{N} = +5.6\text{‰}$, $\delta^{13}\text{C} = -12.6\text{‰}$) and Chicken Feather Standard (CFS; $\delta^{34}\text{S} = -3.8\text{‰}$) were analyzed along with the fish muscle as laboratory calibration standards. Analytical precision of replicated controls was within ± 0.3‰.

Isotope δ values are presented as the per mil (‰) differences from international reference materials ($\delta^{15}\text{N}$: atmospheric N_2 ; $\delta^{13}\text{C}$: Vienna Pee Dee Belemnite (VPDB); $\delta^{34}\text{S}$: Vienna Cañon Diablo Trolite (VCDT)) where $\delta\text{X} = R_A/R_S - 1$; and where X is one of ^{15}N , ^{34}S or ^{13}C , R_A was the ratio of the heavy to light isotope (example: $^{13}\text{C}/^{12}\text{C}$) in the sample and R_S was the ratio in the international standard (Fry, 1988).

2.2. Trace element concentrations

Fish muscle samples reserved for Al, As, Cd, Cu, Fe, Mn and Se analyses were oven dried at 60 °C, homogenized (Vander Zanden et al., 2005), and digested in high purity nitric acid (HNO_3) and 30% v/v analytical grade hydrogen peroxide (H_2O_2). Method blanks, DORM-2 (Dogfish Muscle Certified Reference Material for Trace Metals; National

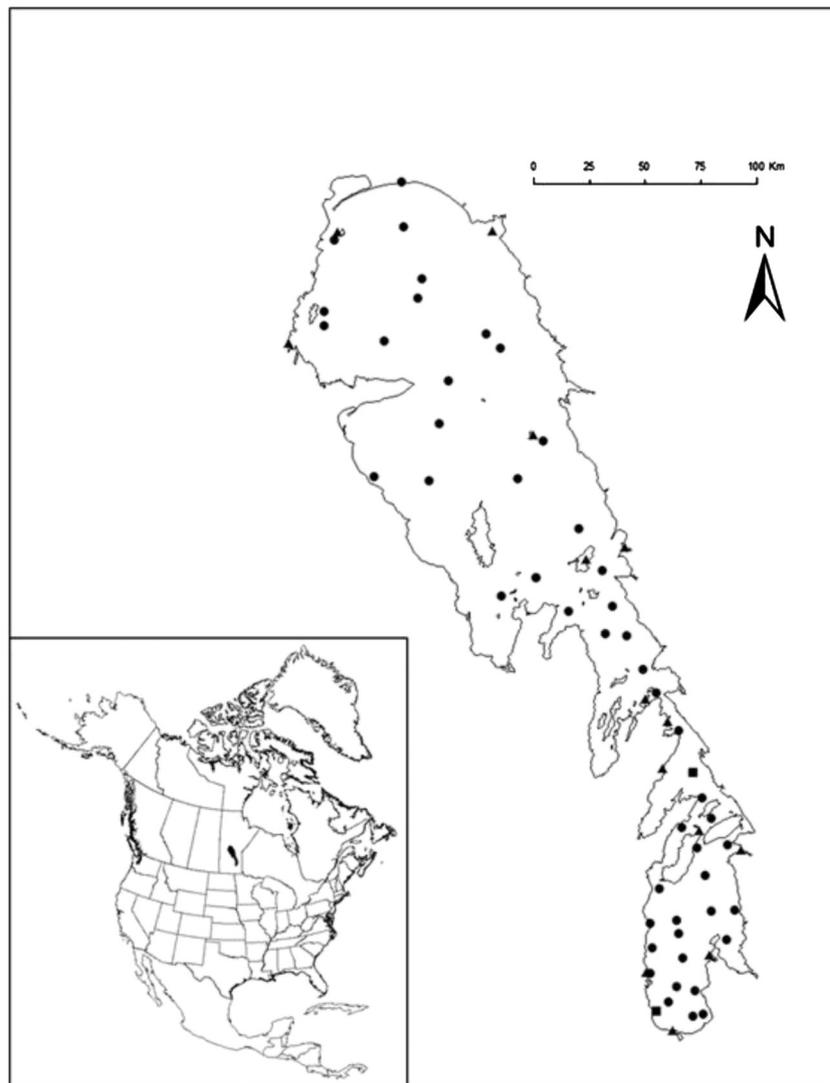


Fig. 1. Lake Winnipeg, Manitoba, Canada. Fish specimens were collected during the ice-free seasons of 2009 and 2010 (beam trawls: closed circles; gillnets: closed triangles) and the winter of 2009–2010 (gillnets: closed squares). The basins are separated by the narrows one-third of the way north.

Research Council Canada; Ottawa, ON) and duplicate samples were included in each digestion batch at a ratio of one blank, reference and duplicate for every ten unknowns. Elemental concentrations were quantified via Inductively Coupled Plasma Mass Spectrometry (ICP-MS) at the Toxicology Centre, University of Saskatchewan, Saskatoon, Canada (Phibbs et al., 2011). Acceptable elemental recoveries from DORM-2 and coefficients of variation (CV) on duplicate samples were $\pm 20\%$ and $\pm 15\%$, respectively (Brumbaugh et al., 2005). Limits of detection (LoD) for Al, As, Cd, Cu, Fe, Mn and Se were better than $0.9 \mu\text{g/g}$, $1.1 \times 10^{-3} \mu\text{g/g}$, $1.8 \times 10^{-3} \mu\text{g/g}$, $5.0 \times 10^{-3} \mu\text{g/g}$, $0.1 \mu\text{g/g}$, 1.3×10^{-3} and $8.0 \times 10^{-4} \mu\text{g/g}$ dry weight (dw), respectively.

2.3. Mercury

Fresh, rather than dried, fish muscle samples were microwave digested (US EPA, 1996) and analyzed for total Hg (hereafter “Hg”) via cold-vapor atomic fluorescence spectrometry (CVAFS; Baker et al., 2004). Digestions and analyses were conducted at the Saskatchewan Research Council (SRC) Environmental Analytical Laboratory in Saskatoon, Canada. Blanks, reference materials and duplicates were analyzed along with the muscle samples. Recoveries of Hg from DORM-2 were between 85 and 100%, except for two low recoveries of 75% and 78%. The CV on duplicate samples was within 15%, and the limit of detection (LoD) was

$0.01 \mu\text{g/g}$ wet weight (ww). Since all other concentrations are presented on a dw basis, percent moisture data for each muscle sample were used to convert the ww Hg data to units of $\mu\text{g/g}$ dw.

2.4. Statistical analyses

Baseline $\delta^{15}\text{N}$, $\delta^{13}\text{C}$ and $\delta^{34}\text{S}$ values of nutrients, primary producers, and primary consumers differ between Lake Winnipeg’s north and south basins (Hobson et al., 2012; Ofukany, 2012). The spatial isotopic heterogeneity identified for zooplankton ($\delta^{15}\text{N}_{\text{Zoop}}$), dissolved sulfate ($\delta^{34}\text{S}_{\text{DS}}$) and dissolved inorganic carbon (DIC; $\delta^{13}\text{C}_{\text{DIC}}$) may confound the interpretation of isotopic data for higher consumers (Ofukany, 2012; Post, 2002). In order to compensate for this variability, isotope values for fish muscle were normalized according to:

$$\delta X_{\text{Corrected}} = \delta X_{\text{Fish}} - \delta X_{\text{Baseline}} \quad (2)$$

where X was one of ^{15}N , ^{13}C or ^{34}S , and $\delta X_{\text{Baseline}}$ was the mean $\delta^{15}\text{N}_{\text{Zoop}}$ (north = $+3.4 \pm 1.9\%$, $n = 31$; south = $+9.6 \pm 2.8\%$, $n = 27$), $\delta^{13}\text{C}_{\text{DIC}}$ (north = $-3.4 \pm 2.0\%$, $n = 31$; south = $-8.4 \pm 1.0\%$, $n = 30$) or $\delta^{34}\text{S}_{\text{DS}}$ (north = $-4.7 \pm 3.1\%$, $n = 30$; south = $-8.2 \pm 2.5\%$, $n = 29$) value for the basin where the fish was captured (Hobson et al., 2012; Post, 2002). Mean $\delta^{15}\text{N}_{\text{Zoop}}$, $\delta^{13}\text{C}_{\text{DIC}}$ and $\delta^{34}\text{S}_{\text{DS}}$ values for each

basin were used in Eq. (2) in order to compensate for within-basin fish movements. The potential seasonal migration of fishes between the two basins was assumed to be negligible (Hobson et al., 2012). Details regarding the measurement of $\delta^{15}\text{N}_{\text{Zoop}}$, $\delta^{13}\text{C}_{\text{DIC}}$ and $\delta^{34}\text{S}_{\text{DS}}$ in zooplankton, DIC and dissolved sulfate respectively, are presented by Ofukany (2012).

Trophic position (TP) was determined for each species within each sub-basin, and was used to compare $\delta^{15}\text{N}$ data across systems. Estimates of TP were calculated from $\delta^{15}\text{N}$ values in fish muscle and zooplankton ($\delta^{15}\text{N}_{\text{Zoop}}$), where zooplankton was assumed to occupy a TP = 2 and all subsequent fish TPs were derived from:

$$\text{TP}_{\text{Fish}} = 2 + \left(\delta^{15}\text{N}_{\text{Fish}} - \delta^{15}\text{N}_{\text{Zoop}} \right) / 3.4\%$$

where $\delta^{15}\text{N}_{\text{Fish}}$ was the nitrogen isotope value for fish tissue and the dietary isotopic discrimination factor ($\Delta^{15}\text{N}$) at each trophic step was 3.4‰ (Gewurtz et al., 2006; Jardine et al., 2006).

A general linear model with Tukey's Honest Significant Difference (HSD) for multiple comparisons was used to determine whether isotopic data for north and south basin fishes came from the same distribution. A plot of species- and basin-specific $\delta^{15}\text{N}$, $\delta^{13}\text{C}$ and $\delta^{34}\text{S}$ values was used to examine fish community structure in terms of three-dimensional isotopic space. Concentrations of trace elements were log-transformed prior to all statistical analyses to meet assumptions of normality based on Shapiro–Wilks' test (Shapiro and Wilk, 1965).

We used general linear models to describe trace element concentrations in fish muscle based on species, sampling period, dietary parameters ($\delta^{15}\text{N}$, $\delta^{13}\text{C}$, and $\delta^{34}\text{S}$) and fish size (FL), and Tukey's Honest Significant Differences for post-hoc comparisons. Statistical analyses were conducted in R 3.0.2 (R Development Core Team, 2013), and effects were considered significant when $p < 0.05$.

3. Results

3.1. Stable isotope values

There were significant differences in stable isotope δ values among species (Wilks' $\lambda = 0.396$, $F_{48,4132} = 31.49$, $p < 0.0001$), sub-basins (Wilks' $\lambda = 0.407$, $F_{3,1389} = 675.87$, $p < 0.0001$), and the species \times basin interaction was also significant (Wilks' $\lambda = 0.849$, $F_{33,4093} = 7.07$, $p < 0.0001$). Univariate analyses showed that $\delta^{13}\text{C}$ varied by species ($F_{16,1704} = 41.18$, $p < 0.0001$), basin ($F_{1,1704} = 986.62$, $p < 0.0001$), and the species \times basin interaction was significant ($F_{12,1704} = 3.42$, $p < 0.0001$). Within each species, $\delta^{13}\text{C}$ was consistently lower in north basin fish, and the difference was significant for all species (Tukey's HSD, $p = < 0.0001$ – 0.028) except for longnose sucker, which did not differ in $\delta^{13}\text{C}$ between basins (Tukey's HSD, $p = 0.433$), this was likely a function of the small sample size and high variance in south basin samples (Table 1).

North basin goldeye had the lowest $\delta^{13}\text{C}$ values of all species, which indicated they were the most pelagic fishes in either basin (France, 1995). Carbon isotope values for north basin white bass and emerald shiner were also among the lowest. For longnose sucker and white sucker collected from the north basin, $\delta^{13}\text{C}$ values in the muscle were representative of relatively benthivorous feeding strategies (Stewart and Watkinson, 2004). Lake whitefish also had $\delta^{13}\text{C}$ profiles which fell toward the benthic end of the $\delta^{13}\text{C}$ spectrum; however their marginally higher $\delta^{15}\text{N}$ (Table 1) may be indicative of some piscivory.

The distribution of south basin fishes along the pelagic–benthic $\delta^{13}\text{C}$ gradient was different from the north basin. Rainbow smelt and troutperch, rather than goldeye, white bass or emerald shiner, had the lowest $\delta^{13}\text{C}$ values of any south basin species (Table 1). Longnose sucker collected from the south basin had the highest $\delta^{13}\text{C}$ values (Table 1, Fig. 2); however values for south basin freshwater drum and walleye were also among the largest.

Fish muscle $\delta^{15}\text{N}$ also differed significantly among species ($F_{16,1656} = 45.43$, $p < 0.0001$) and basin ($F_{1,1656} = 1894.62$, $p < 0.0001$); the species \times basin interaction was also significant ($F_{11,1656} = 10.61$, $p < 0.0001$). Within-species differences were identified where fish data were available for both basins; $\delta^{15}\text{N}$ was consistently higher in the north basin for all species (Tukey's HSD, $p = < 0.0001$ – 0.001), except white bass (Tukey's HSD, $p = 0.37$) and longnose sucker (Tukey's HSD, $p = 0.99$).

Walleye and sauger had the highest $\delta^{15}\text{N}$ of north basin fish (Table 1). Within the south basin fish community, white bass, northern pike and sauger had the highest $\delta^{15}\text{N}$. South basin emerald shiner and cisco occupied the lowest TPs identified for all Lake Winnipeg fishes ($\text{TP}_{\text{EMSH}} = 3.1 \pm 0.4$; $\text{TP}_{\text{CISC}} = 3.2 \pm 0.5$; Table 1, Fig. 2).

The $\delta^{34}\text{S}$ values of fish muscle showed the same overall pattern, differing among species ($F_{16,1426} = 14.86$, $p < 0.0001$), basins ($F_{1,1426} = 306.49$, $p < 0.0001$), and with a significant species \times basin interaction ($F_{12,1426} = 4.94$, $p < 0.0001$). Within each species, $\delta^{34}\text{S}$ was higher in south basin cisco, emerald shiner, freshwater drum, rainbow smelt, sauger, and white bass ($p < 0.001$), while there was no difference in $\delta^{34}\text{S}$ in longnose sucker, northern pike, white sucker, or yellow perch from the two basins ($p > 0.11$). The overlap in $\delta^{34}\text{S}$ values ($\delta^{34}\text{S}_{\text{overlap}} \approx 8.0\%$) between the north and south basin fish communities was much greater than that of $\delta^{13}\text{C}$ ($\delta^{13}\text{C}_{\text{overlap}} \approx 4.5\%$).

Troutperch collected from the lake's north basin had the lowest $\delta^{34}\text{S}$ values of all species, which would indicate that this group was most reliant on detrital sources of sulfate (Croisette et al., 2009). Troutperch from the south basin also had much lower $\delta^{34}\text{S}$ values than other south basin fishes, except for longnose sucker (Table 1). The highest mean $\delta^{34}\text{S}$ value for north basin fishes was that of ninespine stickleback; however, δ values for northern pike, walleye, rainbow smelt, and white sucker were statistically similar (Tukey's HSD, $p \leq 0.05$; Table 1). These species were most likely to have rejected detrital sources of sulfate (see below; Ahlgren, 1996; Croisette et al., 2009). Rainbow smelt had the greatest $\delta^{34}\text{S}$ values of all south basin fishes (Table 1, Fig. 2).

3.2. Trace element concentrations

Of all the elements measured in fish muscle, Fe was found in the greatest concentrations, and ranged from 3.06 to 60.43 $\mu\text{g/g dw}$ in south basin walleye and goldeye, respectively. Concentrations of Al were consistently below the LoD, except in one north basin rainbow smelt (Table 2; 0.77 $\mu\text{g/g dw}$, $n = 15$). Therefore, no models for Al were evaluated. Three and six of the Hg concentrations measured in north basin ciscoes ($n = 28$) and rainbow smelt ($n = 26$), respectively, were below the LoD. A single south basin yellow perch ($n = 9$) also fell below the Hg LoD. These specimens were assigned Hg concentrations equal to half the LoD. All concentrations of As, Cd, Fe, Mn and Se were above their respective LoDs.

We found species to be the most frequent significant predictor of trace element concentrations (13/15 elements), followed by fork length (7/15 elements), $\delta^{13}\text{C}$ (3/15 elements), $\delta^{15}\text{N}$ (1/15 elements), and $\delta^{34}\text{S}$ (1/15 elements; Table 3). Only species influenced the As concentrations in fish muscle. Lake whitefish, rainbow smelt, and freshwater drum had the highest concentrations, while northern pike, sauger, white bass, emerald shiner, and goldeye had the lowest As concentrations (Table 3). Cadmium was also influenced only by species, with rainbow smelt, and emerald shiner with the highest concentrations, and walleye, northern pike, cisco, freshwater drum, goldeye, and lake whitefish with lower concentrations (Table 3).

Concentrations of Co in fish muscle were negatively related to fork length ($\beta \pm \text{SE} = -0.007 \pm 0.002$). Among species, the highest concentrations were in cisco, goldeye, freshwater drum, rainbow smelt and lake whitefish, and the lowest in northern pike and walleye, though the differences among species were small (Table 3). Cu was negatively related to $\delta^{13}\text{C}$ ($\beta = -0.047 \pm 0.017$), $\delta^{15}\text{N}$ ($\beta = -0.027 \pm 0.025$), and fork length ($\beta = -0.002 \pm 0.001$). Emerald shiner and longnose sucker had

Table 1

Mean \pm SD (n) $\delta^{13}\text{C}$, $\delta^{15}\text{N}$, and $\delta^{34}\text{S}$ for the fishes of Lake Winnipeg's north and south basins. Stable isotope data for muscle is baseline corrected (Eq. (2)). Isotopic δ value units are per mil (‰) relative to a primary reference standard.

Species ^a	Abbreviation	Basin	$\delta^{13}\text{C}$	$\delta^{15}\text{N}$	$\delta^{34}\text{S}$
Burbot	BURB	South	-18.1 ± 0.9 (5)	$+6.6 \pm 2.2$ (5)	-0.6 ± 1.2 (5)
Cisco	CISC	North	-23.0 ± 1.5 (101)	$+8.1 \pm 1.7$ (95)	-2.2 ± 1.4 (65)
		South	-18.5 ± 2.5 (167)	$+3.9 \pm 1.7$ (163)	$+0.4 \pm 1.7$ (127)
Emerald shiner	EMSH	North	-23.3 ± 1.3 (106)	$+7.7 \pm 1.3$ (107)	-2.5 ± 1.6 (102)
		South	-19.4 ± 1.0 (68)	$+3.6 \pm 1.5$ (69)	0.0 ± 1.1 (55)
Freshwater drum	FWDR	North	-22.6 ± 0.6 (13)	$+7.9 \pm 0.9$ (12)	-4.0 ± 1.1 (11)
		South	-16.9 ± 3.7 (64)	$+4.8 \pm 1.5$ (60)	-0.8 ± 2.2 (62)
Goldeye	GOLD	North	-24.3 ± 0.9 (4)		
		South	-18.6 ± 3.4 (120)	$+5.6 \pm 1.3$ (119)	-0.1 ± 1.6 (107)
Lake whitefish	LKWF	North	-17.7 ± 4.3 (37)	$+7.7 \pm 1.5$ (36)	-1.8 ± 1.7 (33)
Longnose sucker	LNSC	North	-21.0 ± 0.7 (7)	$+6.7 \pm 0.9$ (7)	-2.9 ± 1.4 (6)
		South	-16.8 ± 4.4 (4)	$+5.4 \pm 0.8$ (4)	-2.6 ± 1.3 (4)
Mooneye	MOON	South	-19.4 ± 0.1 (2)	$+5.7 \pm 1.5$ (2)	-0.3 ± 0.5 (2)
Ninespine stickleback	NNST	North	-22.8 ± 1.2 (18)	$+7.1 \pm 2.0$ (18)	$+0.8 \pm 1.7$ (14)
Northern pike	NRPK	North	-21.7 ± 1.8 (11)	$+9.4 \pm 1.1$ (11)	-0.6 ± 1.2 (11)
		South	-18.4 ± 1.9 (20)	$+6.7 \pm 1.3$ (18)	-0.9 ± 1.4 (15)
Rainbow smelt	RNSM	North	-22.2 ± 0.8 (244)	$+8.1 \pm 1.3$ (235)	-0.9 ± 1.3 (177)
		South	-19.6 ± 0.5 (26)	$+5.6 \pm 1.8$ (26)	$+2.0 \pm 2.4$ (20)
Sauger	SAUG	North	-21.6 ± 1.4 (33)	$+10.8 \pm 1.4$ (33)	-2.1 ± 2.8 (30)
		South	-17.7 ± 1.9 (109)	$+6.6 \pm 1.3$ (104)	-0.0 ± 2.2 (96)
Troutperch	TRPR	North	-22.1 ± 1.1 (19)	$+8.0 \pm 0.9$ (20)	-5.2 ± 1.2 (20)
		South	-19.5 ± 0.4 (31)	$+4.9 \pm 1.1$ (31)	-1.7 ± 1.9 (26)
Walleye	WALL	North	-21.3 ± 1.9 (121)	$+10.4 \pm 1.2$ (118)	-0.5 ± 2.2 (117)
		South	-17.1 ± 2.6 (151)	$+5.5 \pm 1.7$ (146)	$+0.4 \pm 2.1$ (112)
White bass	WHBS	North	-23.5 ± 1.5 (18)	$+8.2 \pm 2.1$ (18)	-2.0 ± 1.7 (18)
		South	-18.0 ± 3.3 (88)	$+7.0 \pm 1.5$ (85)	$+0.1 \pm 1.6$ (82)
White sucker	WHSC	North	-21.3 ± 1.5 (55)	$+6.8 \pm 1.1$ (53)	-1.1 ± 1.4 (49)
		South	-17.5 ± 3.2 (8)	$+4.0 \pm 1.6$ (8)	$+0.7 \pm 2.2$ (8)
Yellow perch	YLPR	North	-21.3 ± 1.4 (29)	$+9.2 \pm 1.8$ (26)	-2.3 ± 1.4 (28)
		South	-18.7 ± 1.3 (55)	$+4.8 \pm 1.2$ (56)	-0.8 ± 1.6 (53)

^a Scientific names: burbot (*Lota lota*); cisco (*Coregonus artedii*); emerald shiner (*Notropis atherinoides*); freshwater drum (*Aplodinotus grunniens*); goldeye (*Hiodon alosoides*); lake whitefish (*Coregonus clupeaformis*); longnose sucker (*Catostomus catostomus*); mooneye (*Hiodon tergisus*); ninespine stickleback (*Pungitius pungitius*); northern pike (*Esox lucius*); rainbow smelt (*Osmerus mordax*); sauger (*Sander canadensis*); troutperch (*Percopsis omiscomaycus*); walleye (*Sander vitreus*); white bass (*Morone chrysops*); white sucker (*Catostomus commersonii*); yellow perch (*Perca flavescens*).

the highest muscle Cu concentrations; sauger, northern pike, walleye, freshwater drum, and lake whitefish had the lowest (Table 3).

Fe was the highest in goldeye, cisco, northern pike, freshwater drum and emerald shiner, and the lowest in rainbow smelt and walleye; no other variables predicted Fe concentrations (Table 3). There was a positive relationship between $\delta^{13}\text{C}$ and Hg in fish muscle ($\beta = 0.021 \pm$

0.021), a positive relationship with fork length ($\beta = 0.006 \pm 0.001$), as well as differences among species. The highest Hg concentrations were in sauger, white bass, northern pike, and walleye, and the lowest in cisco, emerald shiner, and troutperch (Table 3, Fig. 3).

Fork length was negatively related to Mn in the muscle ($\beta = -0.002 \pm 0.001$). Concentrations were highest in longnose sucker and emerald shiner, and lowest in sauger, white bass, walleye, and lake whitefish (Table 3). Molybdenum was negatively related to fork length ($\beta = -0.003 \pm 0.001$), and while there were statistically significant differences among species, the range of values was small (mean concentration of 0.01–0.03 $\mu\text{g/g}$), and did not likely represent any biological significance (Table 3). Similarly, Ni concentrations differed by species, but only the pairwise comparison of goldeye and cisco was statistically significant.

Sr differed only among species; the highest concentrations were observed in white bass, goldeye, and freshwater drum, and the lowest in northern pike, rainbow smelt, walleye, sauger, cisco, and lake whitefish (Table 3). Differences in Sr were only found among species, where emerald shiner, goldeye, lake whitefish and rainbow smelt had the highest concentrations, and freshwater drum, sauger, northern pike, and walleye had the lowest (Table 3). Thallium (Tl) was negatively related to fork length ($\beta = -0.017 \pm 0.003$), and although our general linear model detected a difference among sampling periods, this was driven by low sample size in the fall ($n = 2$; all other samples from the summer). Both samples collected in fall had comparatively low Tl concentrations. There were also species differences, with the highest concentrations in rainbow smelt, goldeye, cisco, emerald shiner, and walleye, and the lowest in freshwater drum, lake whitefish, and northern pike (Table 3).

Fork length was a significant, but weak negative predictor of V concentrations ($\beta = -0.0002 \pm 0.0001$), and again there were differences among species. The highest concentrations of V in fish muscle were in

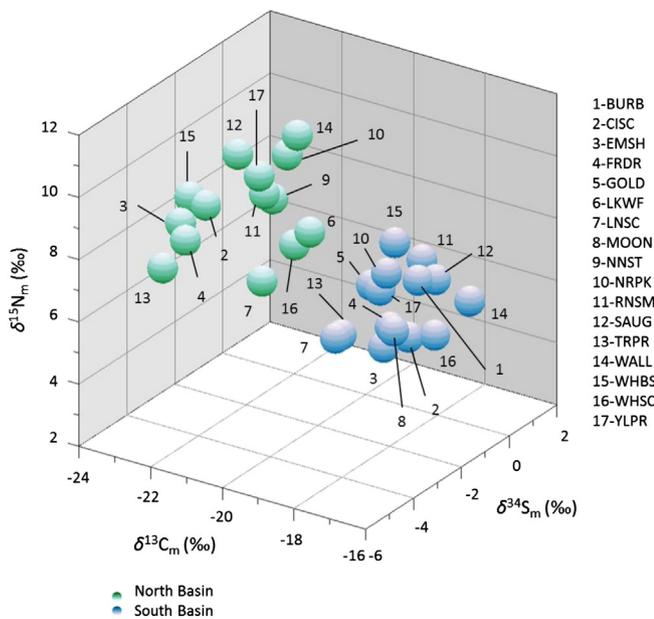


Fig. 2. Isotopic ($\delta^{15}\text{N}$, $\delta^{13}\text{C}$ and $\delta^{34}\text{S}$) food web structure for Lake Winnipeg fishes. Each point represents the mean $\delta^{15}\text{N}$, $\delta^{13}\text{C}$ and $\delta^{34}\text{S}$ values of a given species within either the north or south basin. Species abbreviations as in Table 1.

Table 3
Results of general linear models to describe trace element concentrations in the Lake Winnipeg fish community. Significant terms ($p < 0.05$) are indicated in bold.

Element	log ₁₀ -transformed	Parameter	Degrees of freedom	F-value	p-Value
As	No	Species	7, 46	15.70	<0.001
		Period	2, 46	0.62	0.54
		δ ¹³ C	1, 46	0.02	0.90
		δ ¹⁵ N	1, 46	2.57	0.12
		δ ³⁴ S	1, 46	3.87	0.06
		Fork length	1, 46	1.96	0.17
Cd	Yes	Species	7, 24	4.70	0.002
		Period	2, 24	1.07	0.36
		δ ¹³ C	1, 24	0.10	0.76
		δ ¹⁵ N	1, 24	1.77	0.20
		δ ³⁴ S	1, 24	1.20	0.28
		Fork length	1, 24	0.08	0.78
Co	Yes	Species	7, 30	39.00	<0.001
		Period	2, 30	0.48	0.62
		δ ¹³ C	1, 30	1.09	0.30
		δ ¹⁵ N	1, 30	0.39	0.54
		δ ³⁴ S	1, 30	0.09	0.76
		Fork length	1, 30	17.83	<0.001
Cu	Yes	Species	9, 84	22.30	<0.001
		Period	2, 84	2.29	0.11
		δ ¹³ C	1, 84	11.25	0.001
		δ ¹⁵ N	1, 84	7.92	0.006
		δ ³⁴ S	1, 84	0.66	0.42
		Fork length	1, 84	13.54	<0.001
Fe	Yes	Species	7, 10	3.76	0.029
		Period	2, 10	1.14	0.36
		δ ¹³ C	1, 10	0.11	0.75
		δ ¹⁵ N	1, 10	0.59	0.46
		δ ³⁴ S	1, 10	0.22	0.65
		Fork length	1, 10	0.01	0.93
Hg	Yes	Species	9, 79	17.66	<0.001
		Period	2, 79	1.71	0.19
		δ ¹³ C	1, 79	8.40	0.005
		δ ¹⁵ N	1, 79	0.08	0.78
		δ ³⁴ S	1, 79	0.05	0.82
		Fork length	1, 79	52.66	<0.001
Mn	Yes	Species	9, 46	10.76	<0.001
		Period	2, 46	1.12	0.33
		δ ¹³ C	1, 46	1.26	0.27
		δ ¹⁵ N	1, 46	0.01	0.93
		δ ³⁴ S	1, 46	1.98	0.17
		Fork length	1, 46	10.61	0.002
Mo	Yes	Species	9, 15	5.31	0.002
		Period	2, 15	0.28	0.61
		δ ¹³ C	1, 15	0.32	0.58
		δ ¹⁵ N	1, 15	3.61	0.08
		δ ³⁴ S	1, 15	0.01	0.93
		Fork length	1, 15	9.86	0.007
Ni	Yes	Species	7, 10	4.71	0.014
		Period	2, 10	1.29	0.32
		δ ¹³ C	1, 10	1.39	0.27
		δ ¹⁵ N	1, 10	0.01	0.93
		δ ³⁴ S	1, 10	1.95	0.19
		Fork length	1, 10	0.92	0.36
Pb	Yes	Species	3, 7	0.79	0.54
		Period	–	–	–
		δ ¹³ C	1, 7	0.98	0.36
		δ ¹⁵ N	1, 7	0.44	0.53
		δ ³⁴ S	1, 7	0.08	0.78
		Fork length	1, 7	0.64	0.45
Se	Yes	Species	9, 84	14.80	<0.001
		Period	2, 84	0.58	0.56
		δ ¹³ C	1, 84	0.55	0.46
		δ ¹⁵ N	1, 84	2.41	0.12
		δ ³⁴ S	1, 84	2.81	0.10
		Fork length	1, 84	1.88	0.17
Sr	Yes	Species	9, 14	7.86	<0.001
		Period	2, 14	0.95	0.35
		δ ¹³ C	1, 14	3.44	0.08
		δ ¹⁵ N	1, 14	0.04	0.84
		δ ³⁴ S	1, 14	3.87	0.07
		Fork length	1, 14	1.68	0.22

Table 3 (continued)

Element	log ₁₀ -transformed	Parameter	Degrees of freedom	F-value	p-Value
Tl	Yes	Species	8, 27	9.64	<0.001
		Period	2, 27	7.87	0.009
		δ ¹³ C	1, 27	0.77	0.39
		δ ¹⁵ N	1, 27	3.27	0.08
		δ ³⁴ S	1, 27	0.09	0.77
		Fork length	1, 27	23.34	<0.001
V	No	Species	8, 18	12.80	<0.001
		Period	2, 18	0.41	0.53
		δ ¹³ C	1, 18	9.85	0.006
		δ ¹⁵ N	1, 18	1.95	0.18
		δ ³⁴ S	1, 18	7.48	0.014
		Fork length	1, 18	18.51	<0.001
Zn	No	Species	3, 7	7.34	0.015
		Period	–	–	–
		δ ¹³ C	1, 7	6.41	0.039
		δ ¹⁵ N	1, 7	2.44	0.16
		δ ³⁴ S	1, 7	0.14	0.72
		Fork length	1, 7	2.03	0.20

cisco, freshwater drum, goldeye, and rainbow smelt; the lowest were in northern pike, emerald shiner, sauger and walleye (Table 3). Models predicting Zn and Pb had no significant predictors (Table 3).

4. Discussion

4.1. Food web

The structure of Lake Winnipeg's north and south basin fish communities exhibited a bimodal pattern, similar to that described by Hobson et al. (2012). Yellow perch and northern pike from the south basin of Lake Winnipeg (TP_{YLPR} = 3.4 ± 0.4, n = 56; TP_{NRPK} = 4.0 ± 0.4; n = 20) occupied TPs similar to fishes from Sargent Lake, Michigan (mean TP_{YLPR} = 3.7, n = 33; mean TP_{NRPK} = 4.3, n = 25). North basin perch and pike however, occupied much higher TPs (TP_{YLPR} = 4.9 ± 0.7, n = 29; TP_{NRPK} = 4.8 ± 0.3, n = 11) than both south basin and Sargent Lake fishes (Gorski et al., 2003). This comparison suggests that the structure of the north, rather than south basin food web, deviated from that of other lakes containing perch and pike. The establishment of exotic rainbow smelt within the north basin may have increased the length of the regional food chain (Swanson et al., 2006). However, rainbow smelt collected from the north basin during 2000 to 2002 occupied a TP similar to that of emerald shiner and ciscoes (Gewurtz et al., 2006). Our results for fishes collected in 2009 and 2010 agree with this report (TP_{RNSM} = 4.4 ± 0.4, n = 241; TP_{CISC} = 4.4 ± 0.5, n = 101; TP_{EMSH} = 4.3 ± 0.4, n = 92).

Of the species examined isotopically between 2002 and 2008, walleye were identified as the top predators in each basin (Hobson et al., 2012). Following the inclusion of additional species (BURB, NRPK, SAUG) and larger size classes (WALL, WHBS, YLPR) of fishes in the model, we found that for the north basin, sauger, walleye, yellow perch and northern pike occupied TPs which were higher than other fishes (Fig. 2). North basin sauger and northern pike occupied TPs which were approximately one level above their south basin counterparts; however yellow perch and walleye from the north basin appeared to be feeding at a TP ≈ 1.5 levels higher than those in the south basin (Fig. 2). Previously, we postulated that north basin walleye had larger Δ¹⁵N values relative to south basin walleye, and this was linked to the consumption of protein-rich smelt (Hobson et al., 2012). Here, the relative positions of rainbow smelt and walleye within the north basin food web suggested that a substantial portion of walleye's diet was comprised of smelt (Fig. 2). This trophic linkage was corroborated by historical stomach content analyses (Gewurtz et al., 2006). Alternatively, the increased Δ¹⁵N for north basin walleye and yellow perch may be the result of higher primary production in the north basin. Eutrophication tends to produce conditions favorable to walleye and other percids, with the

increased availability and quality of dietary items generally leading to greater nitrogen isotopic discrimination (Aberle and Malzahn, 2007; Leach et al., 1977). The lack of a similar effect in north basin sauger is likely because environmental conditions within the south, rather than north basin, are more favorable for this species (Johnston et al., 2012).

In lakes of northwestern Ontario, pelagic consumers generally have $\delta^{13}\text{C}$ values between -38 and -26‰ , and consumers classified as characteristically benthivorous tend to have $\delta^{13}\text{C}$ values in the range of -32 to -16‰ (France, 1995). Although fishes from the north and south basins of Lake Winnipeg were within the increasingly benthic range of values ($\delta^{13}\text{C} = -24$ to -16‰ ; Fig. 2) identified for Ontario lakes, the terms “pelagic” and “benthic” were used in the present study to refer to relative position along a $\delta^{13}\text{C}$ gradient.

The greatest relative dependence on pelagic (low $\delta^{13}\text{C}$) versus benthic (high $\delta^{13}\text{C}$) energy sources was observed for white bass and emerald shiner in the north basin (Fig. 2). The results for white bass were corroborated by our earlier isotopic survey of Lake Winnipeg fishes (2002 to 2008), wherein white bass (FL < 350 mm) collected from the north basin had a relatively pelagic feeding strategy (Hobson et al., 2012). The fact that emerald shiner were among fishes with the lowest $\delta^{13}\text{C}$ values in each basin was also not surprising, as fish survey data from the 1990s identified it as an important pelagic species within the lake (Stewart and Watkinson, 2004). The isotopic data for south basin burbot was also similar to historical dietary observations, and indicated that the diet of this group has changed very little over the past 60 years. In the early 1950s, Hewson (1955) conducted controlled diet studies on burbot from Lake Winnipeg's south basin and found that ciscoes were the species' preferred prey. The isotope values here match these earlier findings, given that burbot occupy a TP one step ($\Delta^{15}\text{N} = 3.4\text{‰}$) above ciscoes, $\delta^{13}\text{C}$ values for burbot muscle are greater than mean values for ciscoes by $\approx 1\text{‰}$, and the $\delta^{34}\text{S}$ values of the two species are similar ($\Delta^{34}\text{S} \leq 0.5\text{‰}$).

In recent decades, the density of benthic invertebrates available to benthivores like troutperch, lake whitefish and suckers in Lake Winnipeg has increased substantially (Environment Canada and Manitoba Water Stewardship, 2011). This greater abundance of benthic fauna was reflected in the $\delta^{34}\text{S}$ values of white sucker. White sucker feed

preferentially on benthic prey when plentiful, and only switch to a detritus-based feeding strategy when benthic invertebrate densities are low (Ahlgren, 1996). The relationship between invertebrate density and relative position along the muscle $\delta^{34}\text{S}$ gradient was less straightforward for longnose sucker. Longnose sucker may consume plant matter, and the relatively low $\delta^{34}\text{S}$ values observed for individuals from the south basin may be indicative of an increased reliance on decaying plant materials (Croisette et al., 2009; Stewart and Watkinson, 2004). Troutperch collected throughout the lake had muscle $\delta^{34}\text{S}$ values which indicated frequent consumption of sediment and detritus-based foods. Troutperch are known to have a benthic feeding strategy, and may be consuming a great deal of sediment along with invertebrate prey (Ahlgren, 1996; Stewart and Watkinson, 2004).

4.2. Biomagnification of trace elements

We found no evidence of biomagnification of trace elements in the north and south basin fish communities; only Cu was related to $\delta^{15}\text{N}$, but negatively (Table 3). These findings are similar to those of the food web of Tadenac Lake, Ontario, where Cd, Cu, Fe and Mn did not biomagnify (Wren et al., 1983). Apparent biomagnification effects were seen at finer scales within Lake Winnipeg, namely within walleye and freshwater drum from the lake's south basin. For walleye, the strong positive effect of $\delta^{15}\text{N}$ on Se and Hg concentrations would suggest that as walleye became increasingly more piscivorous, they accumulated greater concentrations of these elements in the muscle. The same may be true for freshwater drum, where individuals who were more dependent on fish (TL ≥ 3) rather than benthic invertebrates (TL ≈ 2) obtained greater amounts of Se from their diet (Stewart and Watkinson, 2004). However, the food web model in Fig. 2 (see also Tables 1 & 2) would suggest that the potential prey items of walleye and drum were unlikely to have exceeded the $3 \mu\text{g/g}$ threshold for diet-induced Se toxicity in fishes (Muscatello and Janz, 2009). Therefore, Se biomagnification within walleye and freshwater drum is unlikely to be a major cause for concern. None of the Se concentrations measured in the muscle approached the United States Environmental Protection Agency's (US EPA) whole-body, chronic limit of $7.91 \mu\text{g/g dw}$ (U.S. EPA, 2004).

We found that total Hg was not related to $\delta^{15}\text{N}$ ($\beta = -0.022 \pm 0.034$), and had a slope considerably lower compared with other studies of temperate freshwater fish (Lavoie et al., 2013). Hg can biomagnify at different rates in benthic and pelagic systems (Borgà et al., 2012), and both were present to some degree in our system. Based on a recent meta-analysis (Lavoie et al., 2013), we would have expected a significantly greater slope estimate (for fish a temperate nutrient-rich freshwater system, around $\beta = 0.16$), but the small range of trophic levels relative to the range of Hg, differences in the degree of specialization among species, and possible movement of fish between basins would mask the Hg– $\delta^{15}\text{N}$ relationship.

Of the walleye from the south basin that were analyzed for Hg ($n = 41$), four exceeded the $1 \mu\text{g/g fw}$ recommended guideline for fish-eating wildlife (Ethier et al., 2008) and two exceeded the limit for commercial sale in Canada (Bodaly et al., 2007). The Hg concentrations we measured were fairly similar to those reported for walleye in other systems. The mean concentration for south basin walleye ($0.76 \pm 0.77 \mu\text{g/g dw}$, $n = 41$) was on the lower end of the range reported for specimens of a similar size that had been collected from lakes in Minnesota and Wisconsin (0.85 to $2.61 \mu\text{g/g dw}$, $n = 79$) (Rolfus et al., 2008).

4.3. Effects of fish size

We found moderate evidence for growth dilution of Co, Cu, Mn, Mo, Tl and V concentrations in fish muscle. As expected, larger fish had greater Hg concentrations (Table 3). The frequent formation of algal blooms within the north basin may be the best explanation for this phenomenon (Pickhardt et al., 2002). Increased algal productivity at the base of the food web has led to increases in fish biomass within the

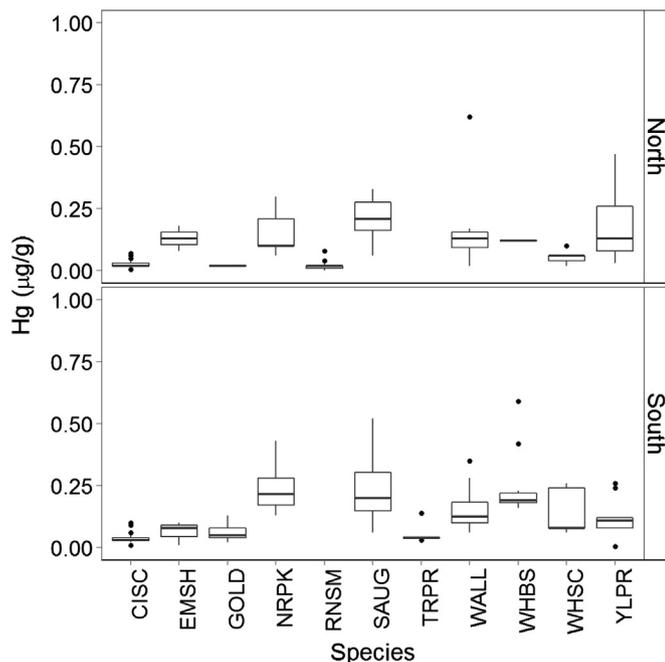


Fig. 3. Total Hg concentrations ($\mu\text{g/g}$) in fish muscle from the north and south basins of Lake Winnipeg. Solid lines are the median, boxes are the first and third quartiles, lines are the 5% and 95% percentiles, and dots are the final outliers. Species abbreviations as in Table 1.

north basin (Environment Canada and Manitoba Water Stewardship, 2011). If the concentrations of trace elements available for uptake by forage fishes were to remain fairly constant, the increase in fish biomass would ultimately decrease the amounts of those elements per unit of growth in fishes (Gewurtz et al., 2006; Swanson et al., 2006). Growth dilution may be more evident in rapidly-growing forage fishes versus larger, slower growing organisms, since the duration of exposure and accumulation per unit of increased length or weight is much lower in fast-growing species (Newman and Unger, 2003).

4.4. Identifying sources of trace elements in diet

The influence of $\delta^{34}\text{S}$ and $\delta^{13}\text{C}$ on trace element concentrations in fish muscle was largely dependent on species. Hg concentrations tended to increase with increasing dependence on benthic foods (high $\delta^{13}\text{C}$), while Cu and V increased with decreasing $\delta^{13}\text{C}$ (more pelagic foraging). Yellow perch collected from lakes near Clyde Forks, Ontario had muscle Hg concentrations which were negatively correlated with $\delta^{13}\text{C}$ (Ethier et al., 2008), while rainbow smelt and pike which were feeding near the sediment in Tadenac Lake, Ontario, accumulated greater concentrations of metals in tissues than relatively pelagic organisms (Wren et al., 1983). The mean Hg concentration for northern pike collected from Tadenac Lake was 4.04 $\mu\text{g/g dw}$ ($n = 20$), assuming the moisture content of pike muscle was 75% (Gorski et al., 2003; Wren et al., 1983). This is substantially higher than the mean Hg concentration for pike from Lake Winnipeg's south basin ($1.16 \pm 0.50 \mu\text{g/g dw}$, $n = 7$).

It is important to note that there were some exceptions to the community-wide generalizations mentioned above for the south basin fishes. For example, goldeye that fed toward the benthic end of the $\delta^{13}\text{C}$ spectrum had lower Cu and greater Mn concentrations in the muscle than would be expected, given the community-level model. South basin goldeye were more similar to north basin fishes in this regard, since Cu concentrations in north basin emerald shiner, lake whitefish, walleye and northern pike tended to be smaller in individuals that obtained dietary energy from the near the sediment, rather than from the water-column. However, rainbow smelt and yellow perch from the same basin tended to exhibit greater concentrations of Cu in the muscle with increased reliance on benthic carbon sources.

4.5. The concentration–diet disconnect

Concentrations of many elements were not related to $\delta^{15}\text{N}$, $\delta^{13}\text{C}$, $\delta^{34}\text{S}$, or fish length. For example, we expected to see a general decrease in Cd concentrations with increasing TP (Campbell et al., 2005). Despite the apparent disconnect between Cd and TP, concentrations were sufficiently low so as not to pose a significant risk to human consumers. The EU's maximum level (ML) of Cd allowed in commercial fishes is 0.05 $\mu\text{g/g FW}$ (Ciardullo et al., 2008). None of the concentrations measured in fish muscle exceeded this limit (Table 2), and the maximum concentration, which was measured in a north basin rainbow smelt, was equal to 0.17 $\mu\text{g/g dw}$, or approximately 0.03 $\mu\text{g/g fw}$.

The mean Hg concentration in the muscle of northern pike (mean \pm SD = $0.69 \pm 0.43 \mu\text{g/g dw}$, $n = 5$) was approximately one-fifth that of northern pike collected from Tadenac Lake, Ontario (Wren et al., 1983), and slightly below the range of concentrations reported for northern pike collected from 17 lakes throughout Minnesota and Wisconsin (0.79 to 5.15 $\mu\text{g/g dw}$, $n = 401$) (Rolfus et al., 2008). Selenium concentrations in the muscle of north basin northern pike (Table 2) were well below the whole-body guidelines for fish in the United States (U.S. EPA, 2004). However, Se may be a potential problem for white bass populations within Lake Winnipeg. Of the $n = 3$ north basin white bass for which there were Se concentration data, two (4.43 and 5.61 $\mu\text{g/g dw}$) approached the US EPA threshold of 5.85 $\mu\text{g/g dw}$, above which further monitoring is recommended (U.S. EPA, 2004), though drawing definitive conclusions is not possible with such a small sample size. A third specimen had a muscle-Se concentration (9.62 $\mu\text{g/g dw}$) which

exceeded the US EPA's 7.91 $\mu\text{g/g dw}$ draft criterion (U.S. EPA, 2004). Unfortunately, due to the small Se sample size for north basin white bass, we were unable to evaluate any potential food web models and identify any diet-related factors which could have contributed to the elevated Se concentrations we observed. Further monitoring of Se concentrations in Lake Winnipeg white bass is recommended.

An important consideration when comparing stable isotope values and elemental concentrations in tissues is their relative periods of integration of information. For the comparison to be informative, the stable isotopes and trace elements must reflect the same processes (e.g., dietary intake) over the same period (Bond, 2010). A direct comparison also assumes negligible (or similar) routing of particular isotopes or elements to different macromolecules (e.g., carbohydrates vs. lipids) (Boecklen et al., 2011; Voigt et al., 2008; Wolf et al., 2009). Stable isotope values in fish muscle integrate diet over roughly 75–100 days prior to sampling (e.g., Buchheister and Latour, 2010). However, Hg and other bioaccumulating trace elements tend to increase with age (and size) (Qian et al., 2001; Somers and Jackson, 1993), especially when older fish feed on prey with higher Hg concentrations; accumulation is ultimately related to metabolism and energetics (Trudel and Rasmussen, 2006). While using stable isotope values to predict trace element concentrations in aquatic systems is a common practice (Jardine et al., 2006; Lavoie et al., 2013), recent evidence suggests that residency of trace elements in fish muscle, and Hg in particular, could be much longer than C and N typically used in isotopic assays (Madenjian et al., 2012; Van Walleghem et al., 2007, 2013). Alternatively, if the assayed tissue provides isotopic information consistent with long periods of integration (i.e. in cases where diet or location of feeding changes little over an annual cycle), then the isotopic information would be an appropriate indicator of those processes (e.g. biomagnification) influencing trace element dynamics.

5. Conclusions

This study is the first extensive investigation into the influence of food web structure and diet on Hg and other trace elements measured in the muscle of Lake Winnipeg fishes. Isotopic food web data ($\delta^{15}\text{N}$, $\delta^{13}\text{C}$ and $\delta^{34}\text{S}$) indicated that fishes in the lake's north basin generally had lower $\delta^{13}\text{C}$ and $\delta^{34}\text{S}$ values, but greater $\delta^{15}\text{N}$ or TP values, than fishes from the south basin, similar to our 2002–2008 study (Hobson et al., 2012) and further validate our earlier suggestion that the north and south basins of Lake Winnipeg be considered as separate systems when examining food web structure. In terms of trace element trophodynamics, species was often the strongest modulator of concentrations at the community-wide level, and concentrations were rarely related to trophic parameters.

Isotopic studies of the trophic interactions among organisms in Lake Winnipeg's north and south basins may serve as a valuable tool for monitoring effects to the fish community and other high-level consumers (e.g., avian and mammalian piscivores) that may result from changes in nutrient loading and sources, introduction of invasive species, and climate change.

Although trace element concentrations in fish tissues collected from Lake Winnipeg did not appear to be related to trophic parameters, ongoing, concurrent sampling of fish for stable isotopic studies and metals analysis may be informative, given larger samples sizes are used. The extent of the commercial fishery on Lake Winnipeg, coupled with the lack of information regarding sources, accumulation and biomagnification of metals in fish tissues, emphasizes the ongoing need to monitor the health of the Lake Winnipeg fish community.

Supplementary data to this article can be found online at <http://dx.doi.org/10.1016/j.scitotenv.2014.07.125>.

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Supplemental Material

Table S1. Fork lengths (FL; mm) of fishes collected from Lake Winnipeg during 2009 and 2010.

Species	n	Basin	Mean \pm SD	Minimum FL	Maximum FL
Burbot	5	South	563 \pm 124	362	696
Cisco	101	North	127 \pm 45	46	258
	169	South	171 \pm 72	54	413
Emerald Shiner	92	North	63 \pm 11	33	85
	60	South	66 \pm 11	33	86
Freshwater Drum	13	North	256 \pm 151	50	460
	64	South	288 \pm 75	21	514
Goldeye	120	South	181 \pm 81	58	313
Lake Whitefish	36	North	320 \pm 122	84	520
Longnose Sucker	7	North	409 \pm 38	332	449
	4	South	356 \pm 25	330	382

Mooneye	2	South	64 ± 9	57	70
Ninespine Stickleback	18	North	44 ± 6	32	55
Northern Pike	11	North	537 ± 115	340	692
	20	South	591 ± 162	79	784
Rainbow Smelt	241	North	91 ± 22	39	206
	26	South	64 ± 16	35	118
Sauger	34	North	311 ± 64	172	406
	111	South	287 ± 60	38	422
Troutperch	20	North	75 ± 15	50	110
	31	South	61 ± 18	36	100
Walleye	124	North	363 ± 85	52	552
	152	South	311 ± 121	25	664
White Bass	18	North	107 ± 103	25	350
	90	South	234 ± 101	30	432
White Sucker	55	North	387 ± 61	262	530
	8	South	369 ± 101	272	531

Yellow Perch	29	North	199 ± 77	31	304
	56	South	154 ± 72	37	256
