

# Spatial and temporal food web dynamics of a contaminated Lake Ontario embayment, Hamilton Harbour

by

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## Abstract

Hamilton Harbour, a semi-enclosed bay located at the western end of Lake Ontario, is listed as one of the most polluted systems in the Great Lakes. Anthropogenic influences such as four wastewater treatment plants, two steel mills and shoreline development have lead to degradation of this system. A Remedial Action Plan is in place to clean up the harbour by 2015. This study examined the food web dynamics of Hamilton Harbour including 21 species of fish, benthic invertebrates, plankton and macrophytes. Using carbon and nitrogen stable isotopes spatial and seasonal variability throughout the harbour was examined. Zooplankton and phytoplankton collected at three different sites in the harbour showed no significant difference spatially but did show seasonal trends, reaching the highest nitrogen values in early summer. Benthic invertebrates, when observed in  $\delta^{13}\text{C}$ :  $\delta^{15}\text{N}$  biplots, group together by sampling site in each season. Seasonally benthic invertebrates acquire higher nitrogen signatures in summer then decrease in fall at all sites. The fish community in the harbour do not have spatially distinct isotope signatures. Seasonally nitrogen signatures increased at all sites while carbon signatures remained between -25 ‰ and -26 ‰. Overall the plankton and benthic invertebrate nitrogen isotope signatures are higher than the fishes. This indicates that there is a recent change in nutrient source. The likely candidate for nutrient input is an anthropogenic source, such as the wastewater treatment plants discharging into the harbour. Isotope signatures show large variation in fish species collected indicating that the fishes are omnivore generalists that take advantage of available food sources throughout the harbour. Further remediation work, such as habitat modifications, can now be tailored towards generalist omnivores that move throughout the harbour.

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# Chapter 1

## 1.1 General Introduction

The Great Lakes basin drains water from eight states and one province, including many large cities. Habitat destruction, pollution, diversion of water supply, aquaculture, invasive species, sewage and wastewater discharge have combined to significantly affect Great Lakes ecosystems to the point where remediation is necessary in many localities (Beeton 2002, Leach et al. 1999). While the cause and effect of many impacts are now known, much remains to be learned about ecosystems adversely affected by human activities. In particular, appropriate understanding of existing trophic connectivity and the feeding relationships among taxa in ecosystems targeted for remediation would provide useful information for remediative planning.

The Great Lakes Water Quality Agreement was established to restore and maintain Great Lakes aquatic ecosystems (IJC 1999). Under this agreement, 43 Areas of Concern (AOC) were identified as degraded environments. Each AOC develops a Remedial Action Plan (RAP) to address the issues that contribute to, or have caused, the environmental degradation unique to each system, e.g., excess nutrients, loss of fish and wildlife habitat, bacterial contamination. One AOC with a long history of shoreline development and ecosystem degradation is Hamilton Harbour.

Hamilton Harbour (43°14'N, 79°51'W) is a naturally enclosed bay at the western end of Lake Ontario. It is roughly triangular in shape, 8km east to west and 4.8km north to south, with a water volume of  $2.8 \times 10^8 \text{ m}^3$  (MOE 1974). The harbour is connected to Lake Ontario across a natural sandbar by the Burlington Ship Canal (MOE 1974). Grindstone, Red Hill and Spencer creeks drain into the harbour as does Cootes Paradise, a 250 ha marsh at the west end of the harbour. The harbour watershed is approximately 500 km<sup>2</sup>, draining equal amounts of urban, agricultural and rural lands (Hall et al. 2006). The Burlington and Hamilton wastewater treatment plants receive sewage from approximately 700,000 (2006 Census) people, and account for 40 % of the freshwater flow into the

harbour (Harris et al. 1980). In the west Grindstone Creek and Cootes Paradise enter the harbour, each with a wastewater treatment plant upstream. Streams (23 %), storm sewers (7 %) and Cootes Paradise (30 %) contribute the balance of water inflows (Harris et al. 1980). Two steel mills use harbour water for cooling and processing, but there is no net exchange of water. The harbour is also used for shipping and recreational boating.

In concert with efforts to reduce contaminant loadings, ecological research on the harbour has focused on trends in contaminant loadings in sediment or taxa (McCarthy et al. 2004) or on the impacts of harbour degradation on specific groups of organisms, such as larval fishes (Leslie and Timmins 1992) or benthic invertebrates (Johnson and Matheson 1968). To date, no study has attempted to examine the implications of habitat degradation for food web structure in Hamilton Harbour. Food web studies are important to remediation work because understanding the interactions between trophic levels can aid in efforts to re-establish viable ecosystem assemblages. The complexity of trophic interactions and structure in ecosystems can be captured effectively with stable isotope studies that trace energy flow and connectivity between taxa (Post 2002). Food web structure, however, may vary in time and space, with analytical scale holding implications for conclusions about both food web interactions and structure (Warren 1989). Thus, it is important to gather stable isotope information for making inferences about trophic relationships at varying spatial and temporal scales.

Stable isotope analysis is a cost efficient and effective method for analyzing food webs. Traditional food web studies utilize gut content analysis that reveal details about the prior 24-48 hrs of organism forage activities. Stable isotopes can provide a longer term view of the dietary relationships because inferences are based on analyses of body tissues built from diet assimilated over time (Fry and Sherr 1984). The use of stable isotopes is based on consumers incorporating the

isotopic signature of their food into their body tissues in a predictable manner (Rounick and Winterbourn 1986). The ratio of carbon isotopes ( $\delta^{13}\text{C}$ ) changes very little as it moves through the food web (Peterson and Fry 1987), fractionating at about 0.4 ‰ per trophic level (Post 2002). The accumulation of  $\delta^{15}\text{N}$  facilitates the use of nitrogen as a tracer of trophic level in food webs as the heavier  $\delta^{15}\text{N}$  isotope metabolizes faster than the lighter  $\delta^{14}\text{N}$  counterpart, which is excreted (Fry 2006).

Views about past population and community assemblages for the fishes of Hamilton Harbour have been reconstructed mostly from historical commercial and recreational fisheries data (Holmes and Whillans 1984). Reporting bias may explain some of the observed change in the fish community, but the change is undeniable. The Hamilton Harbour fish community has shifted from supporting a viable coldwater fishery for Lake Trout (*Salvelinus namaycush*), Lake Whitefish (*Coregonus clupeaformis*) and Cisco (*C. artedii*), to an ecosystem dominated by Yellow Perch (*Perca flavescens*), White Bass (*Morone chrysops*) and exotic species such as Rainbow Smelt (*Osmerus mordax*) and Common Carp (*Cyprinus carpio*) (Holmes and Whillans 1984). The change in community composition is believed to have resulted largely from habitat loss, poor water quality, contaminants, overfishing and invading species. Restoration efforts in the harbour have focused on the creation of habitat designed to encourage warm water littoral species (Smokorowski et al. 1998). Thus, rock reefs, islands, emergent shoals and ‘log’ cover habitats have been installed in various areas of the littoral zone to attract species with a goal of increasing species richness from 4 species per 100 m electrofishing transect to 6-7 species (Smokorowski et al. 1998). A RAP delisting objective for Hamilton Harbour is to shift the fish community from one indicative of eutrophic conditions, with species such as White Perch (*Morone americana*), Alewife (*Alosa pseudoharengus*), Bullheads (*Ameiurus spp.*) and Carp dominating, to a self sustaining community representative of a mesotrophic

ecosystem with Northern Pike (*Esox lucius*), bass (*Micropertus spp.*), Yellow Perch and sunfish as the dominant species (MOE 1992). From 1990 to 1997 there was an increase in centrarchids, cyprinids, native and turbidity-intolerant species richness, and a decrease in non-indigenous species richness and percent non-indigenous fishes by number (Smokorowski et al. 1998). Since the mid-1990s there has been a significant decline in the percent of specialists and native species caught (Brousseau and Randall 2008). In the early 2000s Spottail Shiners (*Notropis hudsonius*), a turbidity intolerant species, increased in numbers sufficient to suggest improvements in water clarity in the harbour (Brousseau and Randall 2008). While it appears that the fish community is evolving toward the desired assemblage, it has yet to reach the desired composition.

The littoral zone is the most productive area in many ecosystems. With light for photosynthesis plants thrive and are used as cover by benthic invertebrates and spawning and juvenile fishes. The littoral zone, defined as the area with water less than 2 m deep, of Hamilton Harbour is only about 15 % of the total harbour area (Johnson and Matheson 1968), but is probably disproportionately important as fish habitat owing to the low oxygen and high contamination levels in available deep water habitats. Accordingly, habitat degradation was one of the primary concerns for rehabilitating the harbour for fish and wildlife. Creation of new habitat and spawning areas in the littoral zone has benefited many species to date (Smokorowski et al. 1998). Remediation of contaminated sediments and low oxygen levels in the hypolimnion remain as long term goals. Given the importance of the littoral zone as productive habitat and the changes to date brought about in the habitat, the littoral area of the harbour will be an important focus for any study attempting to understand the nature of the trophic linkages that exist among harbour resident species.

The littoral zone of Hamilton Harbour can be divided into four areas on the west, east, south and north shores. Each littoral area is distinct because of local influences. In the west Grindstone

Creek and Cootes Paradise discharge into the harbour. Through the canal connecting Cootes Paradise to the harbour the flow changes direction numerous times a day, mostly as a result of changes in wind direction (Skafel 2000). South of the canal the shore is sheltered from prevailing winds, allowing a dense growth of macrophytes (Leslie and Timmins 1992). Within this section of the harbour over 50 shoreline configurations of wetlands, beaches, reefs and spawning beds that sum to 2 km<sup>2</sup> of fish habitat were constructed in 1993 (City of Hamilton).

Along the north shore the marina basin near La Salle Park is sheltered from harsh wave action by floating breakwaters. Habitat alterations along the north shore include a rock breakwater sheltering a large spawning reef designed for resident fish species (FWHRP 1998a, Brousseau and Randall 2008). An additional 25 habitat modules, ranging from logs to concrete pipes, were also installed to improve spawning, nursery, forage and cover for bass (*Micropterus spp.*), Walleye (*Sander vitreus*), Channel Catfish (*Ictalurus punctatus*), Pumpkinseed (*Lepomis gibbosus*), Black Crappies (*Pomoxis nigromaculatus*) and bullheads (*Ameiurus spp.*) (FWHRP 1998a). A group of seven emergent shoals were also established to provide additional spawning and feeding areas for migratory birds (FWHRP 1998a, Brousseau and Randall 2008).

The eastern section of the harbour historically consisted of lagoons and a sandy beach heavily influenced by waves. Three islands connected by nine emergent shoals now act as a breakwater for the shore. The lee side of each island is a wetland or mudflat for shorebirds. The windward sides are armourstone that slope into reefs of cobble ideal for Lake Trout and Lake Whitefish spawning. Fish habitat has been randomly integrated along the shore of the islands. The centre island was retrofitted with cormorant nesting platforms (FWHRP 1998b). The littoral zone of the eastern shore is generally < 1 m with a substrate of rubble, gravel and clay (Leslie and Timmins 1992), but the shore south of the Burlington Ship Canal is unprotected and subject to wave action from strong prevailing winds.

The southshore of the harbour consists primarily of industrial piers, with little accessible or suitable fish habitat. Littoral areas along the shore reach depths of 5-13 m, are devoid of vegetation and are heavily contaminated (Leslie and Timmins 1992).

The uniqueness of the anthropogenic stressors in each of the identified areas of the harbour argues for separate assessment of food web structure, as opposed to a single global assessment of the harbour. The thesis that follows, therefore, combines the need to develop more detailed and accurate pictures of food web structure in the harbour with the need to consider spatial differences by focusing on the evidence for spatial and temporal differences in the food webs supporting resident fish species in the harbour.

## **Chapter 2**

# **Spatial and temporal food web dynamics of a contaminated Lake Ontario embayment, Hamilton Harbour**

### **2.1 Introduction**

The Great Lakes basin drains water from eight states and one province, including many large cities. Habitat destruction, pollution and diversion of water supply, aquaculture, invasive species, sewage and wastewater discharge have combined to significantly affect Great Lakes ecosystems to the point where remediation is necessary in many localities (Beeton 2002, Leach et al. 1999). While the cause and effect of many impacts are now known, much remains to be learned about the Great Lakes ecosystems that have been adversely affected by human activities. In particular, understanding the existing trophic connectivity and feeding relationships among taxa in the ecosystems targeted for remediation would provide useful information for remediative planning.

The Great Lakes Water Quality Agreement was established to restore and maintain the aquatic ecosystems of the Great Lakes. Under the agreement, 43 Areas of Concern (AOC) were identified as degraded environments. Each AOC develops a Remedial Action Plan (RAP) to address the issues that contribute to and/or have caused the environmental degradation unique to each system, e.g., excess nutrients, loss of fish and wildlife habitat or bacterial contamination. One of the AOCs with a long history of shoreline development and ecosystem degradation is Hamilton Harbour, at the western end of Lake Ontario.

Water quality problems, beach closures and increased industrial development led to community acknowledgment that Hamilton Harbour needed remediation (MOE 1985). Hamilton Harbour was listed as one of the most degraded water bodies in the Great Lakes in 1985 (IJC 1999) and was noted to violate many of the criteria established under the local RAP for a healthy ecosystem (Hall et al. 2006). In particular, issues associated with toxic contamination, poor water

quality, high levels of bacterial contamination, urbanization and land management, shoreline access and aesthetics, and fish and wildlife sustainability were identified as immediate concerns (MOE 1985). Remediation efforts to date have aimed at, and achieved, significant reductions in contaminant concentrations in water, sediments, fishes and birds (MOE 1992).

In concert with efforts to reduce contaminant loadings, ecological research on the harbour was commenced with AOC listing (e.g. Polak and Haffner 1978, MOE 1985, Barica 1989). Past studies have focused on general trends in contaminant loadings in sediment or taxa (McCarthy et al. 2004), or examined the impacts of Harbour degradation on specific groups of organisms such as larval fishes (Leslie and Timmins 1992) or benthic invertebrates (Johnson and Matheson 1968). To date no study has attempted to examine the implications of habitat degradation for food web structure in Hamilton Harbour. The complexity of trophic interactions and structure in ecosystems can be captured effectively with stable isotope studies that trace energy flow and connectivity between taxa (Post 2002). Food web structure, however, may vary in time and space, with analytical scale holding implications for conclusions about both food web interactions and structure (Warren 1989). Thus, it is important to gather the stable isotope information for making inferences about trophic relationships at different spatial and temporal scales.

To better understand the impacts of habitat degradation on Hamilton Harbour, this study uses stable isotope analyses to describe the general spatial and temporal structure of food webs in identifiably different habitat types within the harbour. Analysis will concentrate on resident fish species and test the hypotheses that: (1) trophic position and relationships will be spatially distinct, (2) seasonal shifts will be observed within the food webs for all fish species, and (3) that observed seasonal shifts will be similar in direction and magnitude in all spatially distinct food webs.



## 2.2 Materials and Methods

### 2.2.1 Site

Hamilton Harbour (43°14'N, 79°51'W) is a naturally enclosed bay at the western end of Lake Ontario. It is roughly triangular in shape, 8 km east to west and 4.8 km north to south direction with a water volume of  $2.8 \times 10^8 \text{ m}^3$  (MOE 1974). The harbour is connected to Lake Ontario across a sandbar by the Burlington Ship Canal (MOE 1974). The harbour watershed is approximately 500 km<sup>2</sup> and drains approximately equal amounts of rural, agricultural and urban land (Hall et al. 2006). The Burlington and Hamilton wastewater treatment plants receives sewage from approximately 700,000 people and accounts for 40 % of the freshwater flow into the harbour (Harris et al. 1980). Streams (23 %), storm sewers (7 %) and Cootes Paradise (30 %) contribute the balance of water inflows (Harris et al. 1980). In the west Grindstone Creek and Cootes Paradise enter the harbour, each with a wastewater treatment plant upstream. To the south, Red Hill Creek flows into the harbour and two steel mills use harbour water for cooling and processing, but there is no net exchange of water.

Hamilton Harbour was divided into 4 sampling areas; three littoral areas on the west, east and north shores, and one pelagic zone in the centre of the harbour (Figure 1). Each littoral area was considered to be potentially distinct because of local influences. In the west the littoral zone is influenced by wetland mediated discharges from Cootes Paradise. The north shore is exposed to wave action and urban runoff. The eastern shore is exposed to freshwater inputs from Lake Ontario entering via the Burlington Ship Canal. The south shore of the harbour consists primarily of industrial piers with little access or suitable fish habitat and, therefore, was excluded from sampling. The littoral area was more intensively sampled because the profundal pelagic area of the harbour portions of the harbour are known to be anoxic and contain heavily contaminated sediments (Harris et al. 1980, MOE 1992). In addition, fish habitat remediation has exclusively

occurred in the littoral zone. Therefore, to determine the effects of remediation efforts it was deemed prudent to sample where the majority of remediation work has been done.

### **2.2.2 Field sampling**

Plankton samples were collected biweekly at three sites at the west and north littoral zones and the pelagic zone from May 23 to October 25, 2006. Three net hauls (50 cm diameter, 2 m long, 64  $\mu\text{m}$ ), starting from approximately 1 m above the substrate, were taken at each site (del Giorgio and France 1996). Samples were size fractionated within 24 hours of collection using a series of sieves (Grey et al. 2000). The 1 mm mesh removed large detritus, the 153  $\mu\text{m}$  mesh retained zooplankton and the 20  $\mu\text{m}$  mesh collected phytoplankton (Grey et al. 2000) with fractions examined under a microscope to confirm content. Water exchange with Lake Ontario is dependent on wind and seiche effects leading to variable exchange rates likely to bias locally obtained plankton isotopic signatures and, therefore, sampling was not conducted in the east.

Sediment was collected in the east ( $n = 5$ ), north ( $n = 5$ ) and west ( $n = 4$ ) littoral sites in September 2006 via Ekman grabs along the 1.5 m depth contour. The grab was opened from the top and the sediment samples were carefully removed from the top inch of the water-sediment interface. Leaves and twigs were removed prior to freeze drying. All sediment samples were acidified using 1.2N HCl to remove inorganic carbon before being analysed for stable isotope composition (Bunn et al. 1995).

The benthic community was sampled at all littoral (east, north and west) and pelagic (central) sites once in the spring (May-June), summer (July) and fall (August). The pelagic site was sampled using an Ekman grab filtered through a 64  $\mu\text{m}$  sieve. The littoral sites were sampled by hand collecting rocks and washing invertebrates into a bucket. All invertebrate samples were sorted into like groups (e.g., amphipods, oligochaetes, chironomids, etc) and over a period of at

least 24 hrs were maintained in oxygenated water to allow gut clearance (Hamilton et al. 1992). Periphyton was collected by scraping samples off rocks into vials.

Macrophytes and epiphytes were collected in August of 2006. *Cerataophyllum dermersoni*, *Potamogeton richardsonii* and *Elodea canadensis* were collected from boats at the west end of the Harbour. Samples were spun in salad spinners while being rinsed with filtered water to remove epiphytes. *Myriophyllum spicatum* and *Vallisneria americana* were collected around the harbour by Department of Fisheries and Oceans SCUBA divers. Collected material was agitated with filtered water until epiphytic material was suspended (Jones et al. 2000). All evident macroinvertebrates were removed and the macrophytes stored in vials or Whirl Pacs.

Using historical data from Hamilton Harbour surveys and catches at Cootes Paradise Fishway from 1996-2003, fishes were sorted and ranked by abundance, with twenty-one species identified for collection on the basis of past dominance in the fish community (Table 1). Fishes were collected using a variety of methods including: boat electrofishing (100 m transects), trapnets (24 hr sets, 1.8 m house, 44.5 to 63.5 mm mesh), hoopnets (24 hr sets, 1.2 m house, 6.4 mm mesh), gillnets (1.5 to 2 hr sets, 12.7, 19.1 and 25.4 mm mesh), beach seines (30 m transects, 9.1 m long, 3.2 mm mesh), minnow traps (24 hr sets) and trawling (750 to 1500 m transects, 40 and 70 mm mesh on a 6.1 m bottom otter trawl). All sampling was conducted nearshore with the exception of trawling which was completed at depths of 6-21 m. The use of multiple sampling gears was designed to minimize size and habitat selective sampling bias. Fishes were weighed (g) and measured for fork or total length (mm). Total length was used only for fishes with non-forked tails. Specimens were sacrificed with clove oil and transported to the lab on ice in bags, where they were frozen until processing. A 1-2 g sample of dorsal muscle, free of skin, scales and bone was removed for stable isotope analysis. Scales and otoliths were collected for aging. Stomach contents were frozen for future examination.

### 2.2.3 Stable isotope analysis

All collected fish, invertebrate, plant and plankton samples were freeze dried in a ModulyoD-115 freeze drier for a minimum of 48 hrs at -50 °C. Sediment samples were dried at 60°C for several days. Fish samples were ground into a fine powder using a Retsch MM 301 ball mill grinder. All other samples were ground into powder by hand using a mortar and pestle. For stable isotope analysis 1 mg of powder was used for fishes, 0.5 mg of powder for periphyton, macrophytes, phytoplankton and epiphytes and 0.3 mg of powder for benthic invertebrates and zooplankton. Benthic invertebrate and plankton samples included 3-5 individual replicates, as sampled material allowed. Samples were sent to the Environmental Isotope Laboratory at the University of Waterloo. Fish samples were analyzed with an Isochrom continuous flow stable isotope ratio mass spectrometer (GVInstruments / Micromass-UK) coupled to a Carlo Erba elemental analyzer (CHNS-O EA1108 - Italy). All other samples were analyzed with a Delta Plus continuous flow stable isotope ratio mass spectrometer (Thermo Finnigan / Bremen-Germany) coupled to a Carlo Erba elemental analyzer (CHNS-O EA1108 - Italy). By convention, these materials are set to a value of 0 ‰ from which other international standard materials are measured. Results are corrected to nitrogen standards IAEA-N1 and IAEA-N2 and carbon standards IAEA-CH6, EIL-72 and EIL-32. The error for ball-milled standard material is ± 0.2 ‰ for carbon and ± 0.3 ‰ for nitrogen.

Stable isotope ratios are expressed as delta values ( $\delta$ ) and measured as parts per thousand (‰) differences between the isotope ratio of the sample and that of an international standard. The international standard for carbon is carbonate rock from the Peedee Belemnite formation (Craig 1957) and the nitrogen standard is nitrogen gas from the atmosphere (Mariotti 1983). The  $\delta$  ratio is calculated as follows:

$$1) \delta = \left[ \left( \frac{R_{sample}}{R_{standard}} - 1 \right) R_{standard} \right] * 1000$$

where R is the carbon ( $^{13}\text{C}:^{12}\text{C}$ ) or nitrogen ( $^{15}\text{N}:^{14}\text{N}$ ) isotope ratio for the sample or standard.

## 2.2.4 Data analysis

For analytical purposes, data were assigned to one of two groups: fish or lower trophic level. Fish assemblages in each habitat varied (Table 1) and the set of those used for comparisons among sample areas were reduced to include only species commonly caught in all areas as noted in Table 1. For each group, a minimum convex polygon was defined by enclosing an area in  $\delta^{13}\text{C}$ -  $\delta^{15}\text{N}$  bi-plot space that included the mean stable isotope signatures of all member species  $\pm$  one standard deviation (e.g. Layman et al. 2007, Cornwell et al. 2006). The minimum convex polygon is thus the smallest area capable of enclosing the defined points (means  $\pm$  standard deviations) and, thereby, reducing empty space (Cornwell et al. 2006). The area (Equation 2) and geometric centres (Equations 3 and 4), hereafter called the centroid, for the polygons were calculated as:

$$2) \quad A = \frac{1}{2} \sum_{i=0}^{n-1} (x_i y_{i+1} - x_{i+1} y_i)$$

$$3) \quad C_x = \frac{1}{6A} \sum_{i=0}^{n-1} (x_i + x_{i+1})(x_i y_{i+1} - x_{i+1} y_i)$$

$$4) \quad C_y = \frac{1}{6A} \sum_{i=0}^{n-1} (y_i + y_{i+1})(x_i y_{i+1} - x_{i+1} y_i)$$

where  $x$  and  $y$  are the  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values, respectively, of the outermost points of the polygon.

For comparative purposes, data used from the construction of the fish polygon were adjusted for potential trophic fractionation using values given in Post (2002). Data used for the construction of the lower trophic level polygon were not adjusted. Thus overlap between the resulting polygons would indicate heavy utilization of the resources included in the lower trophic polygon by species included in the fish polygon.

To determine the potential contributions of littoral- or pelagic-derived carbon (Equation 5) for fish and benthic invertebrates in the harbour the two-end member mixing model proposed by Vander Zanden and Rasmussen (2001) was used:

$$5) \%_{preya} = (\delta^{13}C_{cons} - \delta^{13}C_{preyb} - 0.4) / (\delta^{13}C_{preya} - \delta^{13}C_{preyb}) * 100$$

$$\%_{preyb} = 100 - \%_{preya}$$

where  $\delta^{13}C_{cons}$  represents the signature of the consumer and  $prey_a$  and  $prey_b$  represent the signatures, respectively, of the littoral and pelagic model end-members. Epiphyton was used as the littoral end-member and phytoplankton as the pelagic end-member. The model also assumes 0.4 ‰ carbon fractionation (Post 2002).

Statistical analyses were completed in JMP 7.0.1 (SAS Institute) with significant results determined at or below the  $\alpha = 0.05$  level. Linear regression was used to analyze seasonal isotopic relationships for plankton with respect to temperature. Analysis of variance (ANOVA) followed by multiple comparison of means using the conservative Tukey-Kramer's HSD *post hoc* test were used to determine differences in carbon or nitrogen signatures among sampling sites or among seasons for sediment, fish, benthos and plankton. Standard t-tests were used to determine the statistical significance of differences between sites for benthic invertebrates when there were only two sampling sites represented. Bartlett's test for homogeneity of variances was used to determine potential differences in the variability of benthic invertebrate and fish isotope signatures among sites. ANOVA and the Tukey-Kramer HSD *post hoc* test were also used to determine isotopic signature differences in macrophyte species as well as differences among isotopic signatures among fish capture methods. Two-way ANOVA was used to examine interactions between sites and seasons for all species of fish with  $n \geq 5$  at all sampling sites. Only five species of fish were considered when examining site-season interactions as a result of low sample sizes ( $n < 5$ ) in at least one season for all other species. Correlations between sites in species use of littoral carbon were tested using the nonparametric Spearman's rank correlation coefficient and differences in the rate of change in littoral carbon use by species was further examined using linear regression and ANCOVA.

## 2.3 Results

### 2.3.1 Plankton

Phytoplankton and zooplankton carbon and nitrogen stable isotope signatures did not differ significantly among sites (ANOVA  $\delta^{13}\text{C}$  phytoplankton  $F_{2,119} = 1.13$ ,  $p = 0.325$ ,  $\delta^{13}\text{C}$  zooplankton  $F_{2,110} = 0.42$ ,  $p = 0.659$ ,  $\delta^{15}\text{N}$  phytoplankton  $F_{2,119} = 1.30$ ,  $p = 0.277$ ,  $\delta^{15}\text{N}$  zooplankton  $F_{2,110} = 0.69$ ,  $p = 0.505$ ). Phytoplankton carbon signatures varied little between seasons, measuring between -29 and -30 ‰ at all sites, whereas zooplankton carbon signatures increased between 5 and 8 ‰ from spring to summer (Figure 2). Nitrogen isotope values peaked at approximately 24 ‰ in early summer at all three sites, and declined to ~15 ‰ until late September when signatures increased again to ~20 ‰. No pervasive relationship between phytoplankton and zooplankton nitrogen signatures and harbour water temperatures (as measured by the National Water Research Institute, Environment Canada, Burlington, ON) (Figure 2) was found (phytoplankton  $r^2 = 0.002$ ,  $n = 35$ ,  $p = 0.821$ , zooplankton  $r^2 = 0.037$ ,  $n = 34$ ,  $p = 0.274$ ). There was an apparent coupling in spring for zooplankton when  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values rose sharply with increases in temperature and  $\delta^{15}\text{N}$  in spring when temperatures and  $\delta^{15}\text{N}$  values rose synchronously.

### 2.3.2 Benthic invertebrates

#### 2.3.2.1 Spatial

Insufficient taxonomic overlap between sites was observed in spring and fall samples to complete statistically robust spatial comparisons as a result of either differences in species assemblages or low sample sizes. Summer stable isotope bi-plots indicated benthic invertebrate signatures grouped by site (Figure 3). Variability in isotope signatures among taxa at each site was similar for nitrogen (Bartlett's  $\chi^2 = 0.03$ ,  $df = 2$ ,  $p = 0.972$ ) but not for carbon (Bartlett's  $\chi^2 = 7.77$ ,  $df = 2$ ,  $p < 0.001$ ). Organisms from the north site tended to have the highest carbon signatures, mean -20.1 ‰, while organisms collected in the east and west ranged from -29 to -21 ‰, with means of -24.1 ‰ and -24.6 ‰ respectively. Pelagic samples consisted only of oligochaetes which had lower

carbon (ANOVA Tukey-Kramer HSD  $F_{3,121} = 58.10$ ,  $p < 0.001$ ) and nitrogen (ANOVA Tukey-Kramer HSD  $F_{3,121} = 119.76$ ,  $p < 0.001$ ) signatures than any of the organisms from the littoral sampling sites and were excluded from further analyses. Samples from the north and west tended to have the highest nitrogen signatures, ranging from 16 to 22 ‰. Amphipod, chironomid, flatworm, isopod, mussel and snail nitrogen signatures were significantly lower in the east than the west (ANOVA Tukey-Kramer HSD amphipod  $F_{2,22} = 48.01$ ,  $p < 0.001$ , chironomid  $F_{2,8} = 5.99$ ,  $p = 0.026$ , isopod  $F_{2,12} = 6.65$ ,  $p = 0.011$ , flatworm  $F_{1,6} = 58.44$ ,  $p = 0.0003$ , mussel  $F_{1,8} = 691.33$ ,  $p < 0.001$ , snail  $F_{1,11} = 23.79$ ,  $p < 0.001$ ).

Mixing model analysis (Post 2002) indicated organisms collected in the north ( $68.4 \pm 10.0$  ‰) sourced significantly more littoral carbon than organisms from the west ( $50.8 \pm 21.1$  ‰) (Table 1, ANOVA Tukey-Kramer HSD  $F_{2,112} = 9.54$ ,  $p < 0.001$ ). There was less variability in the percentage of littoral carbon used by taxa in the north than in the east or west (Bartlett's  $\chi^2 = 12.83$ ,  $df = 2$ ,  $p < 0.001$ ). Oligochaetes collected at the pelagic site were almost entirely dependent on pelagic sources of carbon with littoral carbon used equalling only  $-3.2 \pm 2.7$  ‰.

### 2.3.2.2 Seasonal

Benthic invertebrate nitrogen signatures increased significantly from spring to summer and decreased in the fall, but remained grouped by sampling site (ANOVA Tukey-Kramer HSD  $F_{2,197} = 48.06$ ,  $p < 0.001$ , Figure 4). The east, north and west sites increased in  $\delta^{15}\text{N}$  from spring to summer, respectively, by 5.7 ‰, 5.6 ‰ and 7.1 ‰, then decreased from summer to fall by 1.1 ‰, 3.1 ‰ and 2.3 ‰, respectively.  $\delta^{13}\text{C}$  increased 1.12 ‰, 1.35 ‰ and 0.90 ‰ in the east, north and west, respectively, from spring to summer. The signatures decreased from summer to fall, measuring respectively in the east, north and west, 0.40 ‰, 0.06 ‰ and 0.35 ‰. The pelagic sampling site followed the same seasonal pattern as the littoral sites, but with smaller season to season shifts. Between spring and summer nitrogen and carbon increased 1.25 ‰ and 0.45 ‰.



Nitrogen decreased from summer to fall (0.50 ‰) as in the littoral sites, but carbon continued to increase (0.35 ‰), and did not show the seasonal decline seen in littoral sites.

### 2.3.3 Macrophytes

Macrophytes in the west had significantly higher nitrogen signatures than plants in either the east or north (ANOVA Tukey-Kramer HSD  $F_{2,68} = 23.32$ ,  $p < 0.001$ ) (Figure 5). Carbon signatures were not significantly different among sites (ANOVA Tukey-Kramer HSD  $F_{2,66} = 45.36$ ,  $p = 0.091$ ). *E. canadensis* had a significantly lower carbon signature (ANOVA Tukey-Kramer HSD  $F_{4,64} = 65.73$ ,  $p < 0.001$ ) and higher nitrogen signature (ANOVA Tukey-Kramer HSD  $F_{4,64} = 30.32$ ,  $p < 0.001$ ) compared to other macrophyte species collected in the harbour.

### 2.3.4 Sediment

There were no significant differences in sediment nitrogen signatures among sites (ANOVA  $F_{2,14} = 1.97$ ,  $p = 0.176$ ) with  $\delta^{15}\text{N}$  ranging from 11.8 to 13.6 ‰. Carbon was significantly different in the east and north (ANOVA  $F_{2,14} = 4.82$ ,  $p = 0.026$ ) with  $\delta^{13}\text{C}$  ranging from -25.4 to -26.1 ‰.

### 2.3.5 Fish

#### 2.3.5.1 Spatial

Twenty-one species of fish were collected in the summer of 2006 with 19 species collected in the east and west and 15 species in the north (Figure 6). The  $\delta^{13}\text{C}$ :  $\delta^{15}\text{N}$  biplot for all species collected in the summer shows tight clustering of fishes in the north and west while in the east some species (e.g. Logperch (*Percina caprodes*), Common Carp (*Cyprinus carpio*) and White Sucker (*Catostomus commersonii*)) did not group with the other fish species. Thus, there was greater variability in  $\delta^{15}\text{N}$  of fishes in the east than in the north or west (Bartlett's  $\chi^2 = 7.86$ ,  $df = 2$ ,  $p < 0.001$ ). Mean nitrogen signatures showed no significant differences among sites in spring (ANOVA Tukey-Kramer HSD,  $F_{2,140} = 0.94$ ,  $p = 0.394$ , Figure 7a) and fall (ANOVA Tukey-Kramer HSD,  $F_{2,179} = 0.90$ ,  $p = 0.410$ , Figure 7c), but mean  $\delta^{15}\text{N}$  signatures from the east site

samples were significantly lower in the summer (ANOVA Tukey-Kramer HSD,  $F_{2,145} = 26.78$ ,  $p < 0.001$ , Figure 7b). Mean  $\delta^{13}\text{C}$  signatures in the west were significantly lower when compared to the north and east in the spring (ANOVA Tukey-Kramer HSD,  $F_{2,140} = 9.11$ ,  $p < 0.001$ , Figure 7a), summer (ANOVA Tukey-Kramer HSD,  $F_{2,559} = 17.69$ ,  $p < 0.001$ , Figure 7b) and fall (ANOVA Tukey-Kramer HSD,  $F_{2,179} = 15.59$ ,  $p < 0.001$ , Figure 7c). In all seasons, mean species  $\delta^{13}\text{C}$  signatures in the north and east showed no significant differences (Table 1). When comparing east and west, five species showed a significant increase in nitrogen and six species had lower carbon signatures (Two way ANOVA, see Table 2).

Significant differences in the littoral carbon sourced by fishes in all study sites existed (ANOVA Tukey-Kramer HSD,  $F_{2,733} = 83.12$ ,  $p < 0.001$  Table 1), with the averages for common fish species in the east, west and north being  $59.7 \pm 13.4\%$ ,  $48.6 \pm 11.8\%$  and  $45.8 \pm 11.1\%$ , respectively. A ranking of the species from largest to smallest littoral carbon use (%) at each sampling site showed a gradual, but consistent, shift from littoral to pelagic carbon reliance. The rate of change in littoral carbon use as measured by linear regression was identical in all sites (ANCOVA  $F_{2,36} = 0.63$ ,  $p = 0.541$ ). Regression intercepts, however, differed significantly (ANCOVA  $F_{2,36} = 162.26$ ,  $p < 0.001$ ), with the lowest ranked fish in the east using nearly as much littoral carbon as the highest ranked fish from the north shore (Figure 8). Spearman's rank correlation coefficient analysis indicated no significant correlation between species rankings in the north and east or west ( $p_{\text{north, east}} = 0.131$ ,  $p_{\text{north, west}} = 0.150$ ) while the east and west were significantly correlated ( $p_{\text{east, west}} = 0.029$ ).

Comparison of trawling (pelagic) and littoral fishing methods indicated a significant difference in the mean carbon and nitrogen signatures of fishes caught in the littoral zone versus those captured in offshore trawls (ANOVA Tukey-Kramer HSD,  $\delta^{13}\text{C}$   $F_{4,1138} = 47.09$ ,  $p < 0.001$ ,  $\delta^{15}\text{N}$   $F_{4,1138} = 26.52$ ,  $p < 0.001$ ). Seven fishes were caught in the trawl with the most abundant catches (Alewife (*Alosa pseudoharengus*), Emerald Shiner (*Notropis atherinoides*) and Spottail Shiner (*Notropis hudsonius*)) showing no significant differences in the carbon signatures of

samples caught littorally or in the trawl (ANOVA Tukey-Kramer HSD Alewife  $F_{1,142} = 0.24$ ,  $p = 0.625$ , Emerald Shiner  $F_{3,61} = 1.81$ ,  $p = 0.156$ , Spottail Shiner  $F_{3,48} = 1.36$ ,  $p = 0.267$ ).

### 2.3.5.2 Seasonal

Fish  $\delta^{15}\text{N}$  signatures increased constantly from spring to fall in the east and north, except in the west where the highest nitrogen signatures were recorded in summer (Figure 9). Examining the fish community as a whole with the stable isotope polygon defining fish feeding patterns there is a decrease in polygon size from spring to fall in the east (Table 3). In the west and north, the polygon was smallest in the summer and largest in the spring.

Carbon stable isotope signatures did not change with season for the fish community as a whole (ANOVA Tukey-Kramer HSD,  $F_{2,1100} = 0.86$ ,  $p = 0.424$ ) but when examining species specific trends carbon signatures were highest in the east or north section of the harbour (Table 2). Mean nitrogen signatures for the fish community increased from spring to fall (ANOVA Tukey-Kramer HSD,  $F_{2,1100} = 103.40$ ,  $p < 0.001$ ). Species specific trends shows Alewife (ANOVA Tukey-Kramer HSD,  $F_{2,142} = 77.29$ ,  $p < 0.001$ ) was the only species, of the 5 species found to be common within site and season, to increase from spring to fall with no site-season interaction (Table 2).

A spatial and temporal comparison of the fish (FP) and lower trophic (LTP) food web stable isotope polygons conducted using areal plots and centroids (Figure 10) indicated the LTP increased in size from spring to summer (Table 3), with the range of both nitrogen and carbon increasing. The FP showed carbon signature remained around -25 ‰ for all sites in all seasons, but the included range of nitrogen signatures increased from spring to summer by 2.5 ‰, and from summer to fall by 0.7 ‰. Percent overlap of the FP and LTP increases from spring to fall in the east and west but remains constant in the north (Table 3). As the seasons progress the LTP nitrogen signatures increased and the FP overlapped in the middle carbon and low nitrogen range

of the LTP. By fall the FP was roughly centered in the LTP with the highest percent overlap between the two polygons.

## 2.4 Discussion

Resident fish species in Hamilton Harbour occupy spatially distinct positions within the identified food webs in the harbour and are closely connected with the locally differentiated lower trophic food webs within the harbour. In addition to supporting the hypothesis concerning the spatially distinct nature of food webs within the harbour, data presented here corroborated the hypothesis concerning seasonal shifts within the harbour food webs and the synchronicity among shifts. Results observed here, therefore, are consistent with patterns of spatial heterogeneity found in other ecosystems (e.g. Boon and Bunn 1994, Adlerstein et al. 2002, Fry et al. 1999, 2008) and patterns of seasonal variation thought to be regulated by changes in nutrient source inputs and tissue turn over cycles in resident organisms (O'Reilly et al. 2002, Bearhop et al. 2004).

Seasonal variation of  $\delta^{15}\text{N}$  and  $\delta^{13}\text{C}$  commonly occurs in food webs (Goering et al. 1990, Toda and Wada 1990, Boon and Bunn 1994). Seasonal changes in isotopic signatures were observed at all trophic levels in Hamilton Harbour. These results emphasize that seasonal sampling is important for Hamilton Harbour because incorrect assumptions, such as feeding strategy or trophic position, could be made from sampling at one point in the year. Coulter (1991) found that the clupeid *Stolothrissa* takes several months to a year to integrate diet signatures whereas temporal integration in primary consumers occur faster (Hecky 1991, O'Reilly et al. 2002) with zooplankton reflecting signatures of phytoplankton within the season (Gu et al. 1994, Leggett et al. 1999). This was observed in Hamilton Harbour plankton nitrogen stable isotope signatures with synchronicity of zooplankton and phytoplankton seasonally. An influx of nutrients will be observed in primary producers and consumers sooner than at higher trophic levels (O'Reilly et al. 2002, Bearhop et al. 2004, Carlier et al. 2000). With trophic correction the fish community is feeding on the lower to mid portion of the lower trophic food web. This may not actually reflect that the fish are feeding on the lower to mid portion of the lower trophic food web but that the plankton and benthic invertebrates have increased in nitrogen due to their

quicker tissue turnover while there has not been enough time for fish to turnover their tissues and express the new high nitrogen signature. The data shows that a new nutrient source has become available in the harbour long enough ago that plankton and benthic invertebrates have turned over their tissues but fishes have not. By fall the fishes are feeding in the middle of the lower trophic food web which may be indicative of the fishes nitrogen signatures increasing or the lower trophic level nitrogen signatures decreasing to a level overlapping with the fishes.

Hamilton Harbour has a history of eutrophication due to shoreline development and anthropogenic influences including four wastewater treatment plants that discharge into the harbour. The wastewater treatment plants are spread around the harbour with two at the east end, discharging directly into the harbour, and two at the west end, one passing through a large marsh and another discharging into a stream that eventually enters the harbour. Anthropogenic nitrogen can be distinguished from natural freshwater because of more positive  $\delta^{15}\text{N}$  values (Heaton 1986, Macko and Ostrom 1994, Clark and Fritz 1997). Sewage nitrogen isotopic signatures range from 8 ‰ to 22 ‰ (Heaton 1986, Clark and Fritz 1997). Littoral organisms sampled in the east end of the harbour had lower nitrogen signatures than in the north or west suggesting Lake Ontario, with lower nitrogen isotopic signatures (Leggett et al. 2000), dilutes effluent. No specific sampling of discharge from the wastewater treatment plants was done but given that sewage nitrogen isotopic signatures range from 8-22 ‰ and plankton signatures in the harbour were  $\sim 24$  ‰ wastewater discharge could be seen as a high nitrogen nutrient source. The spring freshet injects freshwater from the surrounding area but as the seasons wear on there is less freshwater entering the system. Wastewater discharges, however, remain at essentially constant volumes. Therefore, the wastewater input, with high nitrogen values, would be accounting for a larger portion of the inputs into the harbour in summer and fall. There is no new nutrient source but increased concentration of high nitrogen wastewater in summer and fall. It appears that localized inputs are circulated throughout the harbour pelagically, but in the littoral zone sedentary organisms, such as benthic invertebrates, show spatially distinct isotopic signatures with organisms collected in the

west and north sites more directly affected by anthropogenic influences having higher nitrogen signatures than in the east.

Fish have been found to exhibit different feeding strategies when collected in sites separated by only a few kilometres (Jennings et al. 1997, Pinnegar and Polunin 2000, Chassot et al. 2008). Plasticity in feeding behaviour allows organisms to take advantage of local food sources. Prey isotope signatures can vary widely even in small systems (Fry et al. 1999, 2000). Spatial differentiation was found for benthic invertebrates at different sampling sites which were reflected by fish isotopic signatures. It does not appear that fish remain at the site in which they feed though. Specialized feeding of fishes on local food webs will be reflected by a narrower range in isotope signatures. Generalists, however, will display a wider range of signatures due to feeding and accumulation of isotopes from several sources (Fry et al. 1999, Bearhop et al. 2004, Fry et al. 2008). Fishes collected in Hamilton Harbour covered a wide range of carbon isotope signatures and have large variation indicating that they are generalists feeding on both littoral sources with high carbon signatures and pelagic sources with low carbon signatures. The benthic invertebrates collected in the north sampling site have a small carbon isotope range but the fishes collected in the north have the same range as fishes in the east and west indicating that the fishes are generalists integrating a diet from around the harbour and taking advantage of any available food source. This is supported by several fish surveys of the harbour (Smokorowski et al. 1998, Brousseau and Randall 2008) which found that fishes categorized as generalists were most abundant. There was no significant difference in nitrogen isotope signatures for fish between sites and almost complete overlap of fish polygons, representing the isotopic range covered by the fish community, between sites. All of these results suggest that fishes in Hamilton Harbour are integrating a diet from around the harbour and likely taking advantage of any available food source at those sites.

Spatial and temporal variation of stable isotopes in food webs is dependent on prey availability, predator diets and predator-prey interactions. Alderstein et al. (2002) found that

spatial and temporal variation in the diet of haddock was related to prey composition. Variation in feeding habits is dependent on spatio-temporal variations in diet (Jiang and Jorgensen 1996, Alderstein et al. 2002). Isotope signatures of the individual fish species in Hamilton Harbour showed variation among season and site sampled but no dependence of one variable on the other. Fishes following a generalist feeding lifestyle would take advantage of abundant food sources as they become available through the season. Bearhop et al. (2004) stated that organisms consuming a constant proportion of prey will show less variation in isotopic signatures than organisms feeding on differing proportions of prey, what is not taken into consideration is the change in isotopic signatures of prey with season. If fish are feeding on only plankton they will reflect the plankton isotopic signature but plankton signatures change throughout the season giving a planktivorous fish a spring, summer and fall signature to reflect (O'Reilly et al. 2002). Given that tissue turnover takes months to a year in fish species the fish may reflect spring signatures by fall and the fall plankton signatures several months later. Collectively fishes at different sites in Hamilton Harbour displayed stable isotope signatures that varied synchronously through the seasons. Given that variation in stable isotope signatures was similar for the fish communities around the harbour it can be concluded that fish diets differ in proportions.

The food web of Hamilton Harbour changes spatially and seasonally. The more sedentary organisms, such as benthic invertebrates, show that local influences are affecting stable isotope signatures spatially. Synchronicity of seasonal shifts was observed among sites for benthic invertebrates, plankton and fish. Fishes appear to be integrating food sources from throughout the harbour but the wide variability in signatures may be due to seasonal isotopic changes in food sources such as benthic invertebrates and plankton. Without seasonal and spatial sampling in the harbour the intricacies of trophic level interactions would be missed. Given that lower trophic levels are impacted spatially by local inputs remediation efforts should focus on reducing the effect of inputs. Ideally there would be isotopic differentiation between piscivore, specialist and generalist fish species but currently there does not appear to be any segregation with all fish



feeding on a variety of food sources. With improved local littoral habitats and benthic invertebrate communities the specialization of fish species to particular feeding regimes may be attained and spatial segregation of fish communities observed.

## 2.5 Tables

Table 1. A complete list of fish and benthic invertebrate samples with the mean % of carbon derived from littoral sources as estimated from a two-end member mixing model for spatially segregated sample sites in Hamilton Harbour: east, north, west and pelagic.

Sample	Scientific name	Site														
		East				North				West				Pelagic		
		Sample size	Mean (%)	Standard deviation		Sample size	Mean (%)	Standard deviation		Sample size	Mean (%)	Standard deviation		Sample size	Mean (%)	Standard deviation
Alewife	<i>Alosa pseudoharengus</i>	12	52.0	15.9		12	33.5	13.9		32	46.3	10.1				
Bluegill	<i>Lepomis macrochirus</i>	31	49.7	8.2		8	38.4	7.9		38	39.8	10.1				
Bluntnose Minnow	<i>Pimephales notatus</i>					3	52.1	5.1								
Bowfin	<i>Amia calva</i>	1	47.6							1	59.5					
Brown Bullhead	<i>Ameiurus nebulosus</i>	10	67.0	10.8		21	42.2	10.0		38	49.7	10.8				
Channel Catfish	<i>Ictalurus punctatus</i>	3	70.0	3.6						10	67.9	5.9				
Common Carp	<i>Cyprinus carpio</i>	17	52.1	11.0						3	44.6	3.6				
Emerald Shiner	<i>Notropis atherinoides</i>	12	51.9	10.5		11	43.0	12.5		20	52.4	13.9				
Gizzard Shad	<i>Dorosoma cepedianum</i>	1	71.2			2	48.0	0.2		11	36.6	8.4				
Green Sunfish	<i>Lepomis cyanellus</i>	10	58.9	8.3		5	54.8	2.8								
Largemouth Bass	<i>Micropertus salmoides</i>	2	70.7	3.8						14	50.3	7.4				
Logperch	<i>Percina caprodes</i>	10	77.9	4.5		4	52.9	0.9		5	56.6	14.7				
Northern Pike	<i>Esox lucius</i>									21	52.4	5.4				
Pumpkinseed	<i>Lepomis gibbosus</i>	11	58.0	15.3		7	44.6	14.1		12	47.7	10.2				
Rock Bass	<i>Ambloplites rupestris</i>	18	60.8	10.5		10	55.2	8.8		4	47.1	4.4				
Round Goby	<i>Neogobius melanostomus</i>	37	62.0	11.5		30	45.1	8.8		25	50.2	12.8				
Spottail Shiner	<i>Notropis hudsonius</i>	8	59.9	11.1		12	45.6	7.5		8	44.7	8.7				
Walleye	<i>Sander vitreus</i>	15	66.0	4.0						4	58.3	4.7				
White Perch	<i>Morone americana</i>	13	48.2	9.7		11	42.9	10.1		14	44.7	8.4				
White Sucker	<i>Catastomus commersonii</i>	7	66.4	10.6		6	45.9	7.4		6	54.7	8.2				
Yellow Perch	<i>Perca flavescens</i>	39	65.1	16.2		27	52.7	10.2		44	50.1	12.9				
Amphipod		10	51.1	5.6		10	67.4	8.3		5	37.0	4.4				
Chironomid		3	66.6	8.8		5	68.0	6.4		3	75.4	8.8				
Flatworm		3	37.6	1.2						5	38.4	2.6				
Isopods		5	67.7	1.9		5	71.7	3.3		5	61.1	6.7				
Leech						10	62.3	5.7		10	49.1	6.3				
Mayfly						5	66.7	1.4								
Mussel		5	34.2	3.2						5	18.0	4.2				
Oligochaetes																
Snail		5	77.9	12.8		8	55.9	13.3		8	55.9	13.3				
										10	-3.2	2.6				

Table 2. Two way ANOVA results for analysis of the interaction between season and site carbon and nitrogen stable isotope values for the most abundant and common fish species in Hamilton Harbour. Seasons or sites not significantly different from one another are denoted with common letters (A, B or C), with A used to indicate the highest value and C the lowest value. Where significant interactions occurred between season and site a dash (-) is entered in the table. All species were collected at all sites, but not in all seasons.

Nitrogen Species	Season			Site			
	Season (p-value)	Spring	Summer	Fall	East (p-value)	North	West
Alewife	0.097	* <0.0001	C	A	0.838	A	A
Bluegill			B	A	*0.009	B	AB
Brown Bullhead	0.128	0.126	A	A	*0.007	B	AB
Emerald Shiner					0.945	A	A
Logperch					0.389	A	A
Pumpkinseed					*0.006	B	A
Round Goby	*0.003	* <0.001	-	-	0.479	-	-
Spottail Shiner					0.366	A	A
White Perch	0.455	0.277	A	A	0.744	A	A
White Sucker					*0.001	B	A
Yellow Perch	*0.020	* <0.001	-	-	*0.045	-	-
All common species	0.133	* <0.001	C	A	0.219	A	A
<b>Carbon</b>							
Alewife	0.348	0.741	A	A	0.570	A	A
Bluegill					*0.009	A	AB
Brown Bullhead	0.338	0.138	A	A	*0.002	A	B
Emerald Shiner					0.712	A	A
Logperch					*0.012	A	A
Pumpkinseed					0.474	A	A
Round Goby	0.544	0.330	A	A	*0.048	A	B
Spottail Shiner					*0.041	A	AB
White Perch	* <0.001	*0.034	-	-	0.207	-	-
White Sucker					0.198	A	A
Yellow Perch	0.446	0.243	A	A	* <0.001	A	B
All common species	*0.038	0.318	-	-	* <0.001	-	-

\* denotes significant p-value

Table 3. Areas of convex polygons describing  $\delta^{13}\text{C}$ :  $\delta^{15}\text{N}$  bi-plot area occupied by sampled fish and lower trophic level organisms. % overlap is the % of the fish polygon area that overlaps with the lower trophic level polygon.

Site	Season	Area		% Overlap
		Fish	Lower Trophic	
East	Spring	36.32	102.37	50.3
	Summer	25.35	103.13	58.6
	Fall	22.15	63.18	95.1
North	Spring	28.50	69.29	55.5
	Summer	26.44	76.51	54.9
	Fall	32.06	55.07	56.4
West	Spring	25.57	47.82	38.0
	Summer	16.43	109.15	100.0
	Fall	16.18	56.52	100.0

## 2.6 Figures

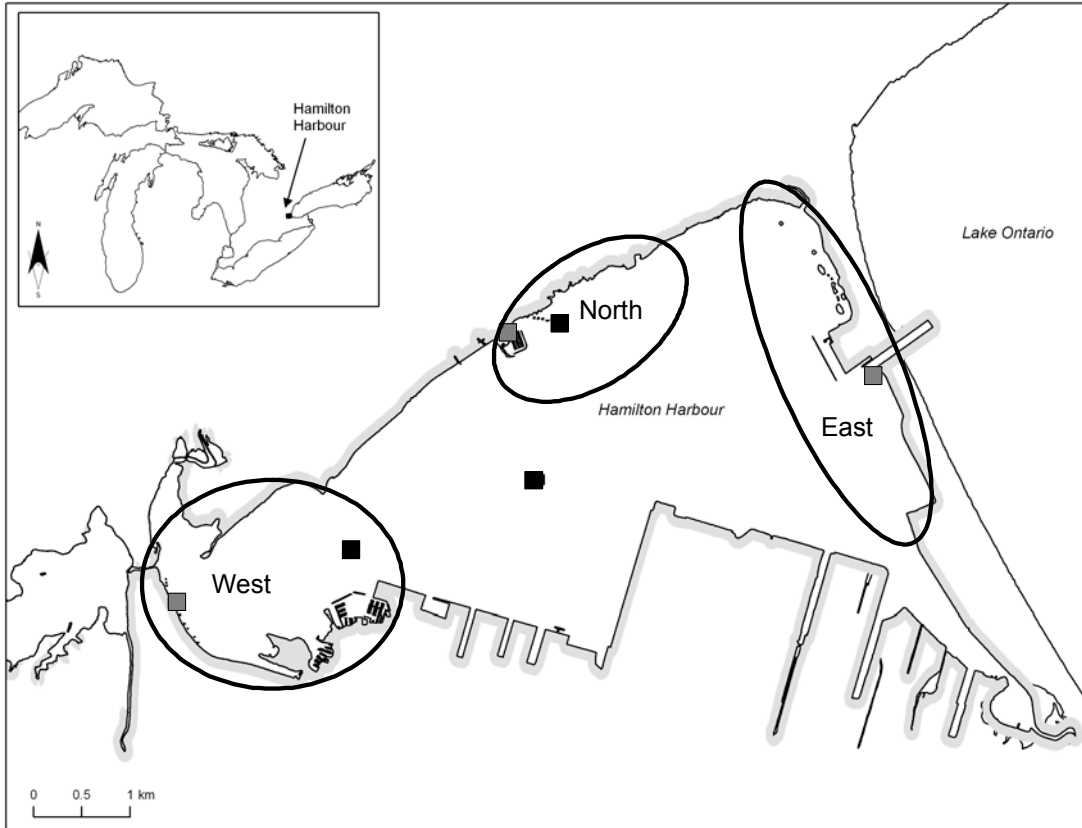


Figure 1. A map of Hamilton Harbour with plankton (black squares), benthic invertebrate (grey squares) and fish (large circles) sampling locations.

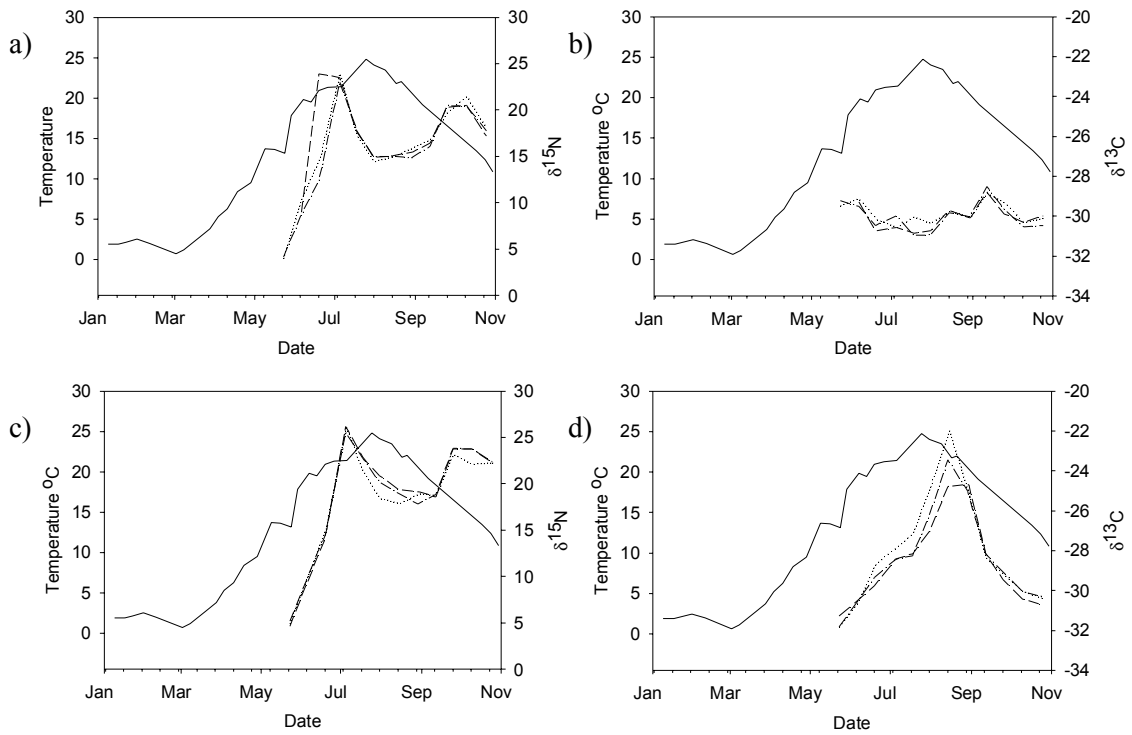


Figure 2. Seasonal variation in phytoplankton (a,b) and zooplankton (c,d) at pelagic (dashed), north (dotted) and west (dot-dashed) sampling sites relative to epilimnetic water temperature (solid).

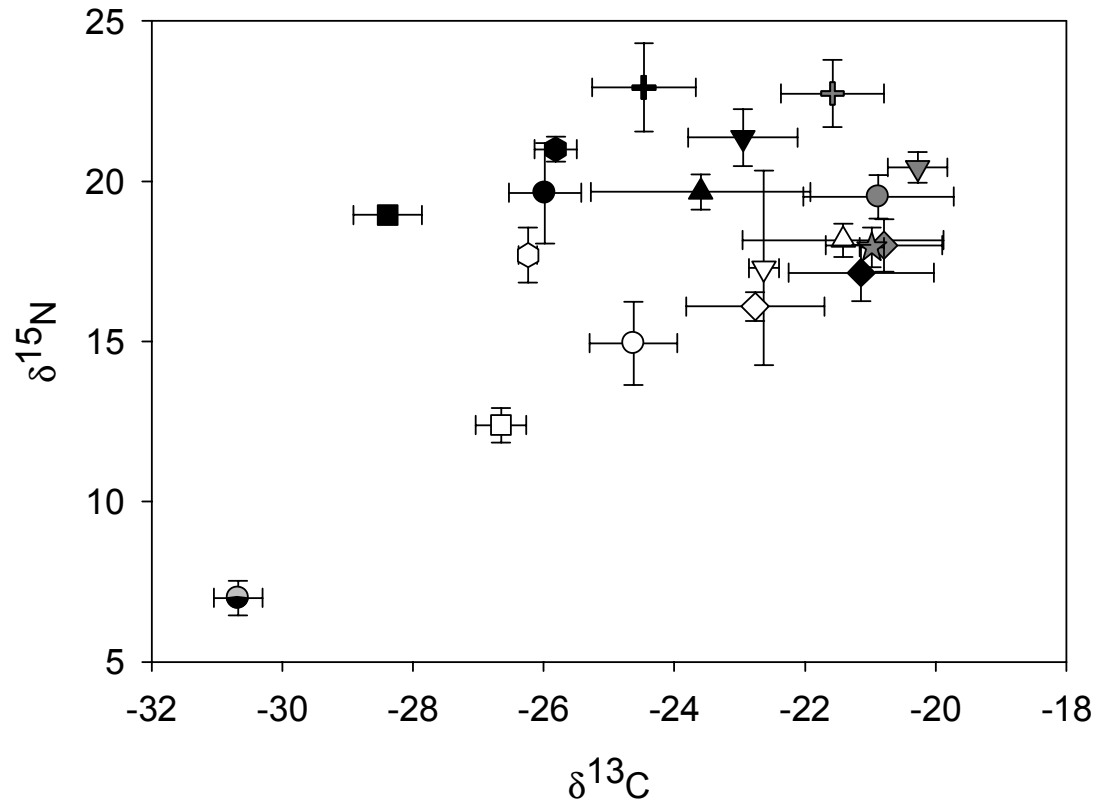


Figure 3. Summer benthic invertebrate taxa (● amphipods, ▼ isopods, ■ mussels, ◆ chironomids, ▲ snails, ● flatworms, † leeches, ★ mayflies, half/half circle oligochaetes) mean stable isotope signatures in sample regions (east- white symbols, north- grey, west- black, pelagic- grey/black) of Hamilton Harbour.

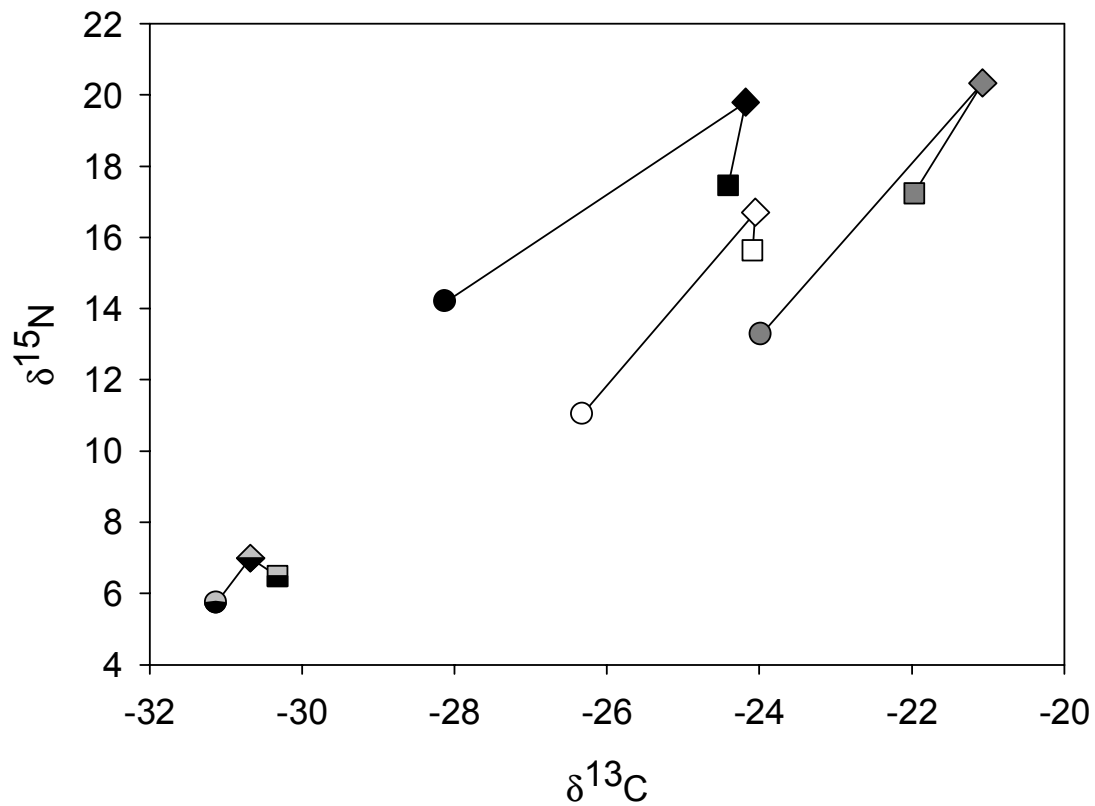


Figure 4. Seasonal and spatial trends observed in benthic invertebrate assemblages as represented by the centroid of the convex polygon defined in  $\delta^{13}\text{C}$ - $\delta^{15}\text{N}$  space for samples obtained for each season (circle-spring, diamond-summer, square-fall) at each site (east-white, north-grey, west-black, pelagic-grey/black).



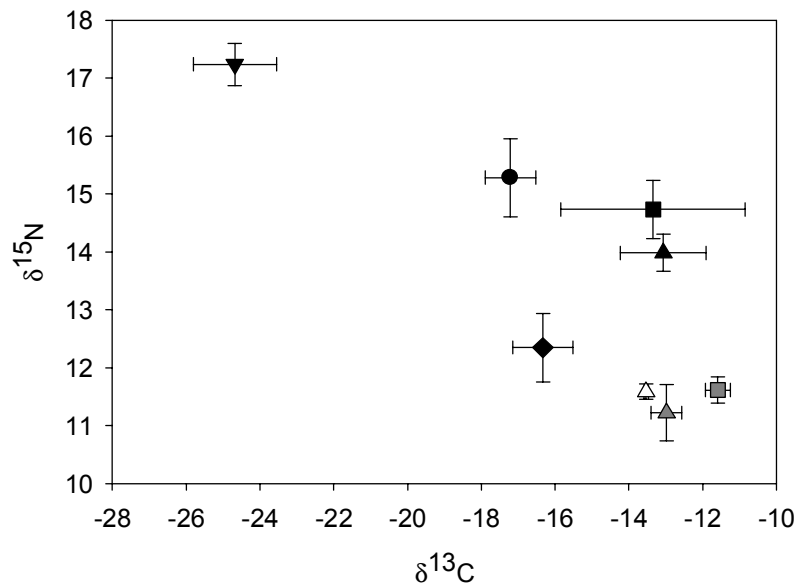


Figure 5. Mean submergent macrophyte (● *Ceratophyllum demersum*, ▼ *Elodea canadensis*, ■ *Myriophyllum spicatum*, ◆ *Potamogeton richardsonii*, ▲ *Vallisneria americana*) stable isotope signatures for samples collected in the east (white), north (grey) and west (black) of Hamilton Harbour.

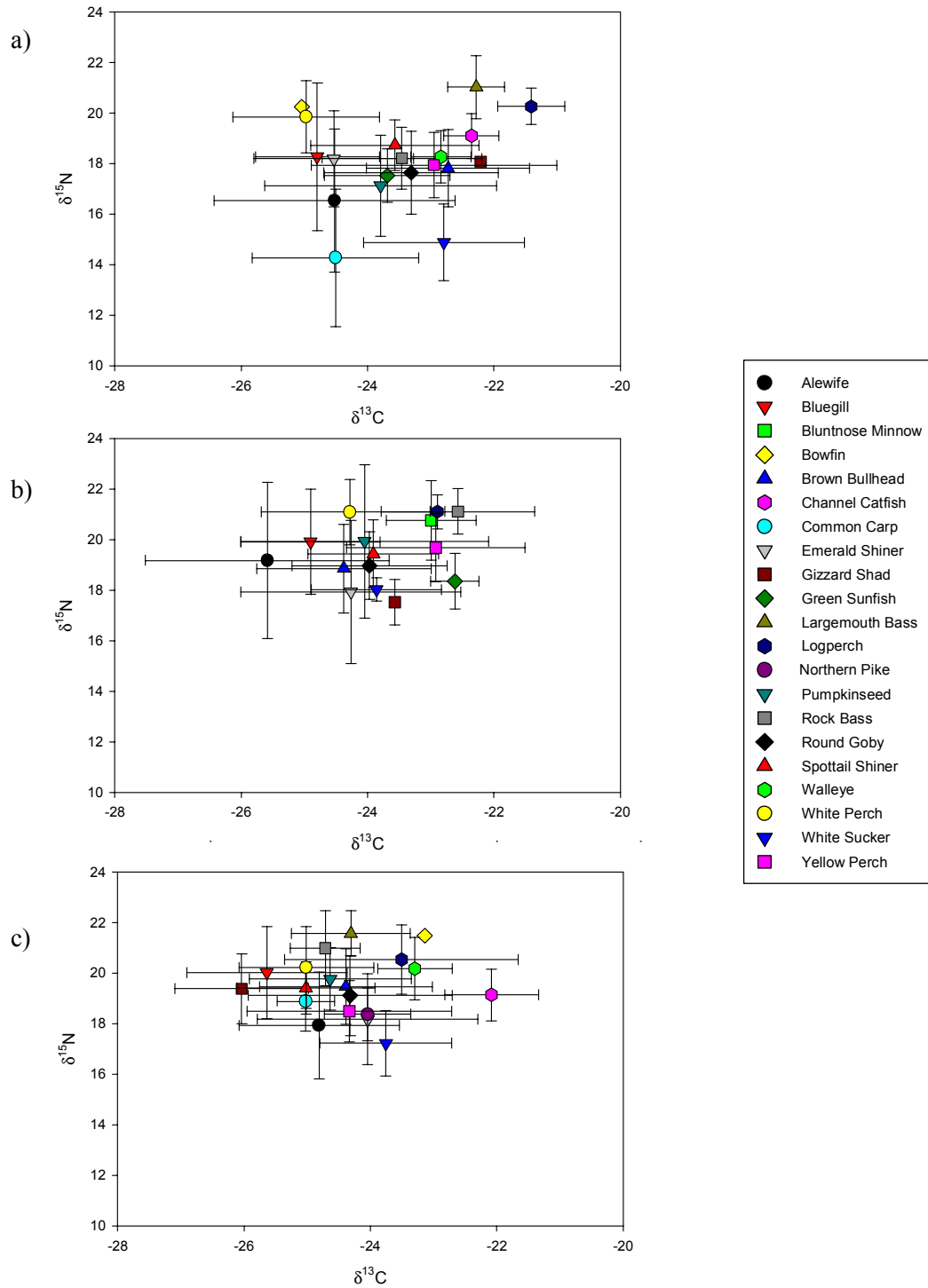


Figure 6. Spatial variation in the stable isotope signatures of fish species caught at sampling locations; east (a), north (b) and west (c) in Hamilton Harbour.

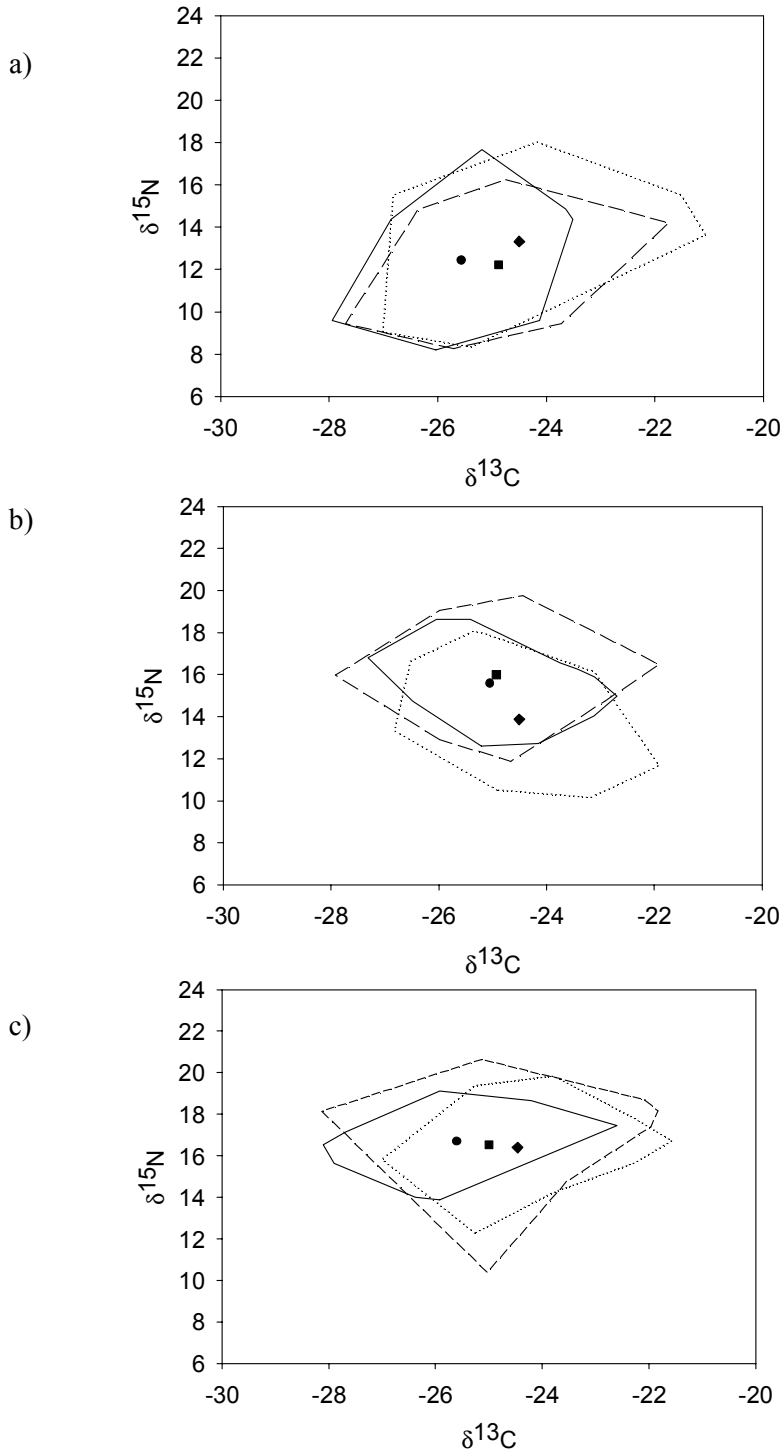


Figure 7. Spatial comparisons of the food web area occupied by common fish species in the spring (a), summer (b) and fall (c) for east (dotted,◆), north (dashed,■) and west (solid,●) sampling sites in Hamilton Harbour.

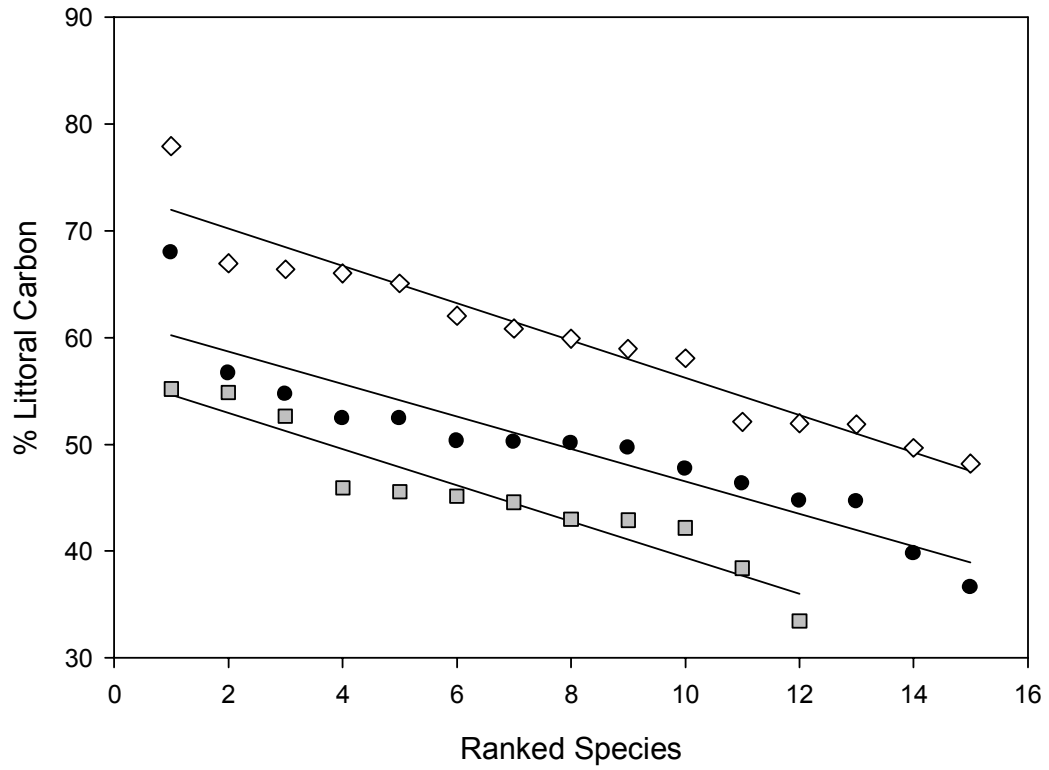


Figure 8. The average % carbon derived from littoral sources for each species of fish at each sampling site (east-white, north- grey, west-black) in Hamilton Harbour. Species were ranked from highest to lowest in each site and solid lines plot significant regression lines for considered variables for each site.

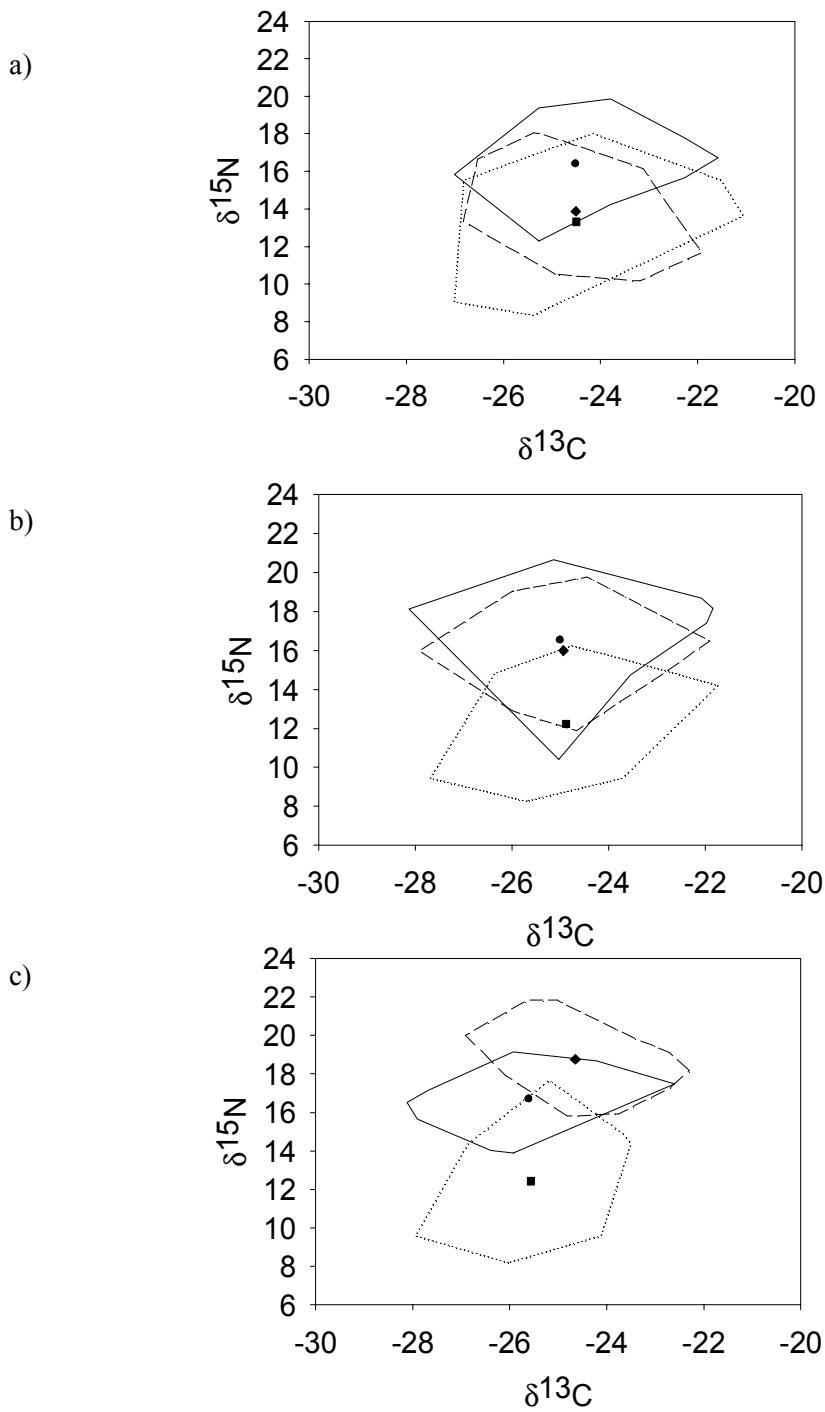


Figure 9. Temporal comparisons of the food web area occupied by common fish species in the spring (dotted, ■), summer (dashed, ◆) and fall (solid, ●) in the east (a), north (b) and west (c) sampling sites in Hamilton Harbour.

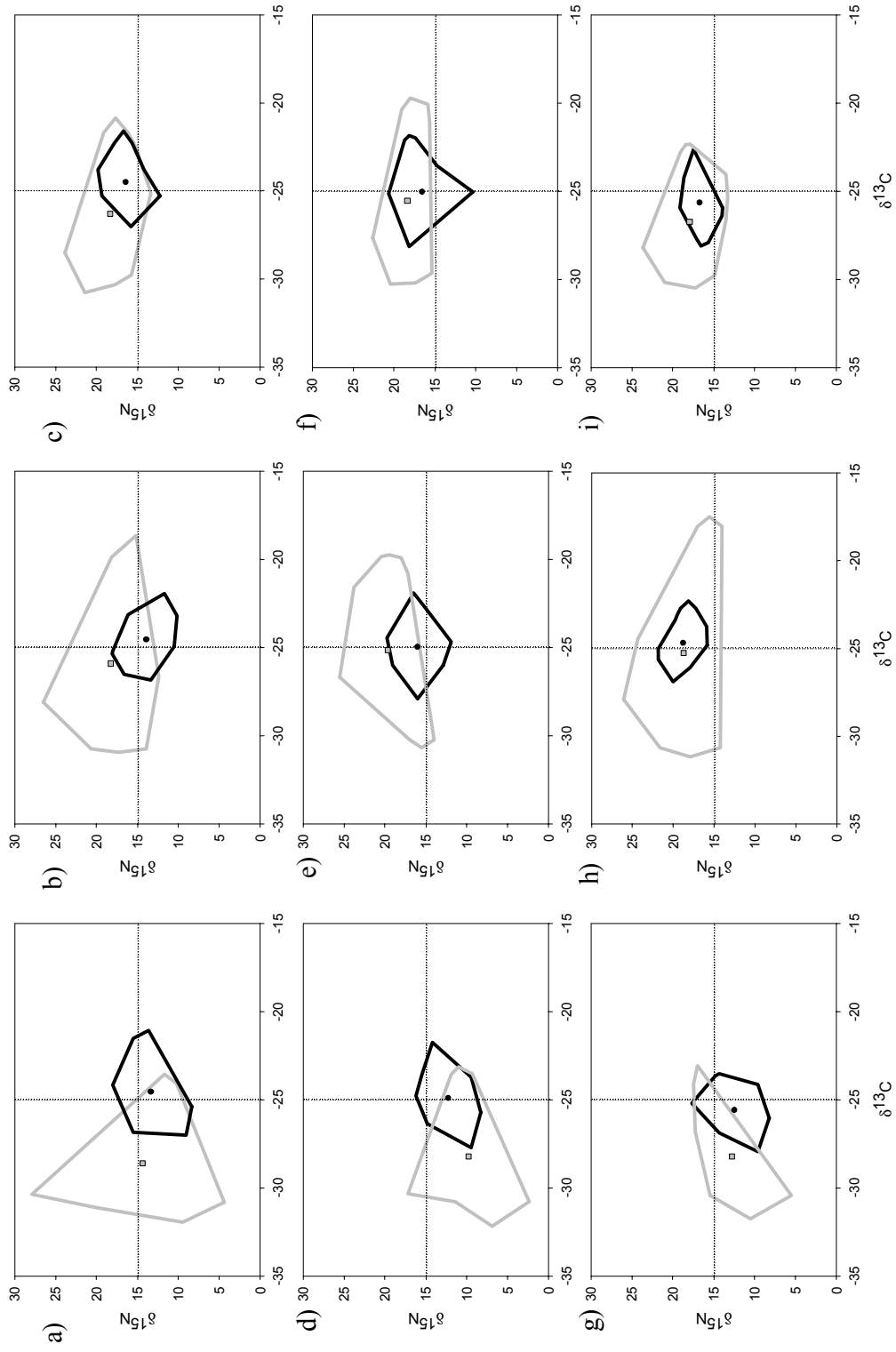


Figure 10. Areal representation of the fish (black, ●) and lower trophic level (grey, □) food web components of Hamilton Harbour in the spring (a,d,g), summer (b,e,h) and fall (c,f,i) in the east (a-c), north (d-f) and west (g-i) sampling sites.

## Chapter 3

### 3.1 General conclusions and future directions

The Remedial Action Plan (RAP) for Hamilton Harbour sets 2015 as the goal for delisting one of the most polluted, degraded systems in the Great Lakes. The vision for the harbour is for it to be a thriving, vibrant centre of the community (MOE 1992). Specific delisting goals were outlined in the RAP and address issues such as loss of fish and wildlife habitat and the establishment of a naturally reproducing warm water fishery. To accomplish these goals a whole ecosystem approach to remediation was undertaken to integrate social, economic and environmental concerns (Hartig and Vallentyne 1989). The whole ecosystem approach can be looked at for the aquatic community as examining all trophic levels and their interactions.

Studies like this one that examine the entire food web and their interactions present opportunities to highlight problems between trophic levels. By undertaking complete food web studies a full view of the ecosystem is shown, that is, top piscivores will be evident by their high nitrogen signatures with prey fish species lower than them and the prey of the fish prey below them. In Hamilton Harbour all of the fish plot, in  $\delta^{13}\text{C}$ :  $\delta^{15}\text{N}$  space, closely together indicating that most species are feeding on similar food sources. It was hypothesized that there would be spatial variation among sites due to habitat restructuring, inputs, and contamination. Fish habitat has been installed in various areas around the harbour, specifically around the areas that were sampled for this project. While some fish species had significantly different isotopic signatures the overall variation of signatures indicated that the fish community as a whole are not staying in one area to feed. By continuing to sample the entire food web and monitor the position of the piscivores, specialists and generalists we can hope to see some differentiation in nitrogen signatures reflected by different feeding regimes with continuing improvements to the harbour. The RAP report outlines the biomass

piscivores, specialists and generalists would ideally represent (MOE 1992). I think that fish categorized as specialists are following a generalist lifestyle in Hamilton Harbour. I believe that the reason for lack of differentiation at present is due to lack of food sources which has forced fish to eat whatever they can, wherever they can. Future remediation efforts might consider focusing on increasing the benthic invertebrate community and creating a pelagic system with plankton that is always consumable.

Seasonal variations of  $\delta^{15}\text{N}$  and  $\delta^{13}\text{C}$  have been reported in other studies and are supported by results of this project (Goering et al. 1990, Toda and Wada 1990). Seasonal variation for  $\delta^{15}\text{N}$  and  $\delta^{13}\text{C}$  can reach 10‰ (Boon and Bunn 1994). Carbon isotopic variations in plankton have been related to changes in species composition (Wong and Sackett 1965), temperature (Sackett et al. 1965) and the  $\delta^{13}\text{C}$  of the carbon being fixed. Size fractionation of plankton samples, as was done in this study, can result in missing shifts in the community structure because all organisms of the same size group together, not sorted by species. Shifts in community structure are observed in carbon isotope signatures (Gearing et al. 1984) and could explain the seasonal changes in the harbour samples. As the temperatures increased and season progressed the dominant species shifted which was reflected in the isotopic signatures. The focus of this project was the fish community and how it interacts with food sources. As a future project a more detailed examination of particular species dynamics for zooplankton and phytoplankton could provide improved insights into productivity and variability spatially in the harbour. Some species of fish may be feeding selectively on particular species of plankton which is currently being overlooked.

The high nitrogen values observed in plankton and benthic invertebrates may be due to the increased volume of wastewater in the harbour in the summer and fall. Sewage has a higher signature than typical freshwater (Heaton 1986, Clark and Fritz 1997) due to preferential loss of  $^{14}\text{N}$  during



denitrification and volatilization of ammonium which are part of secondary and tertiary sewage treatment (McClelland et al. 1997). A more detailed study examining the isotopic signatures and contribution of wastewater discharge from the four treatment plants could indicate if this is the source of enrichment.

The RAP goal of establishing a warm water fishery is foreseeable for the near future but the long term goal of re-establishing a cold water fishery in the harbour is unlikely. The hypolimnion of the harbour becomes anoxic during the summer (MOE 1974) which limits the benthic invertebrate species which can survive. This study observed only oligochaetes in the sludge at the bottom of the harbour. Sediment contamination is one of the major issues preventing delisting of Hamilton Harbour (MOE 1992). Sediment contamination affects the benthic invertebrate population and distribution which in turn can dictate the fish population and distribution in the harbour. It appears that remediation of the harbour must be addressed at every level because the food web is intricate with each trophic level dependant on those below and above it. Continued monitoring of the benthic invertebrate community in the pelagic zone as well as their isotopic signatures and interactions with fish will show improvements to this extremely impacted site.

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