The shifting trophodynamics in four southern Nova Scotia lakes after the introduction of Chain Pickerel (*Esox niger*).

By

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Abstract

Invasive fish species Chain pickerel (*Esox niger*) was first reported within Kejimkujik National Park and Historical Site in 2018. I used stable carbon ($\delta^{13}C$) and nitrogen ($\delta^{15}N$) ratios to assess food web structure and trophodynamics in four four lakes ranging over an invasion spectrum: Loon Lake (first Chain Pickerel report in 2018), Grafton Lake (2019), Big Dam West Lake (2020) and Cobrielle Lake (2021). It was shown that *E. niger* can be grouped into two clusters based on their feeding habits, CP1 and CP2; 4.2cm ≤ TL ≤ 10.9cm and 20.2cm ≤ TL ≤ 58.6cm. Mixing model results indicate CP1 individuals feed primarily on Odonata with a mean dietary proportion of 0.736 ± 0.079. Those assigned to CP2 feed primarily on native fish with a mean dietary proportion of 0.724 ± 0.032. Post-invasion there was a consistent decrease in overall trophic position for fish and Odonata prey items.

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Acknowledgments

This research was carried out in Kespukwitk, one of the seven districts comprising Mi'kma'ki, the traditional and unceded territory of the Mi'kmaq people. Saint Mary's University is located within Sipekne'katik.

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Note: This is a manuscript-style thesis; therefore, similar background information is repeated within the two chapters. Chapter 1 includes background information on the subject matter. Chapter 2 examines the dietary proportions of Chain Pickerel in Nova Scotia freshwater food webs. Chapter 2 was prepared following the Canadian Journal of Fisheries and Aquatic Sciences guidelines and is intended for publication. The authorship for this publication is as follows:

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Chapter 1: A Review of Chain Pickerel (*Esox niger*) and Stable Isotope Analysis

1.1 Description of *Esox niger*

Chain pickerel *Esox. niger* Lesueur, 1818 is a sight-oriented opportunistic predator species (Coffie, 1998; Scott & Crossman, 1973). *E. niger* is in the Esocidae family and thus share similar body shapes and morphologies with its congener species, Northern Pike *Esox lucius* and Grass Pickerel *Esox americanus* (Ross & Brenneman, 2001) (Figure 1). Mature *E. niger* is notably smaller in size when compared to Northern Pike, having an overall smaller total length (Coffie, 1998).). *E. niger* has a distinct lateral chain-link pattern marked by yellowish green areas broken by dark interconnecting lines which gives *E. niger* its common name (Lee et al., 1980; Scott & Crossman, 1973). The chain-link pattern and color acts to camouflage them from prey while in submerging vegetation; it mimics the shadows casted by submerged vegetation in sunlight (Coffie, 1998). There is a prominent dark line below the eye, often referred to as a "tear-drop" (Lee et al., 1980; Scott & Crossman, 1973). They are considered carnivores in general and they most often exhibit qualities of an ambush predator typically occupy shallow lakes and large sluggish rivers containing an abundance of submerged aquatic vegetation and water depths of less than 3m (Coffie, 1998; Scott & Crossman, 1973). Like all *Esox* species, chain pickerel are mostly solitary, spending most of their time hiding motionless in patches of aquatic vegetation (Coffie, 1998; Lee et al., 1980).

When a prey is spotted a chain pickerel will dart quickly towards it (Raney, 1942; Underhill, 1949). Due to their vision-based hunting patterns, chain pickerel are generally more successful at hunting in clear lakes as opposed to brown lakes, due to increased visibility (Hoyle

& Lake, 2011). A series of sensitive proprioceptor nerve endings running along the locomotive muscles, located laterally along the exterior of the skin near the lateral line, allow *E. niger* to remain motionless while they hunt (Ono, 1982). They are also be used to detect the motion of nearby fish as these nerves are sensitive to the pressure waves created by the movement of other fish (Ono, 1982). The undersurface of the lower jaw is pierced by 5 pores on each side (sometimes 4 or 6) (Hoyle & Lake, 2011; Scott & Crossman, 1973). These submandibular pores are filled with fine hairs that are used to detect disturbances in water caused by the movement of prey (Hoyle and Lake, 2011). In short, *E. niger* are a highly adaptive predatory fish and are often considered a top predator within their native and introduced range (Scott & Crossman, 1973).

1.2 The Ontogeny & Reproduction

Spawning takes place over a course of 7 to 10 days during the Spring soon after the ice leaves lakes and rivers, typically in late March or April (Coffie, 1998; Miller, 1962). There is evidence that *E. niger* also spawns in the Fall, from the end of September to early October (Miller, 1962; Scott & Crossman, 1973). No nests are built and there is no parental care of the eggs (Coffie, 1998; Lee et al., 1980). The eggs are yellow, about 2 mm in diameter, and have an adhesive outer membrane that allows them to stick to submerged surfaces (Coffie, 1998; Lee et al., 1980). A single female may spawn several times over the course of one or two days, laying in total between 6,000 to 8,000 eggs (Coffie, 1998).

Fertilized eggs hatch within 6 to 12 days, depending on the water temperature, with eggs hatching faster in warmer temperatures than in colder temperatures (Coffie, 1998). After hatching, the fry secrete an adhesive substance that allows them to stick to submerged vegetation and substrates (Underhill, 1949). The fry feed on their yolk sack for about a week after hatching

(Coffie, 1998). After about a week, the jaw develops and the fry begin feeding on plankton almost exclusively, they are planktivorous at about 10 mm in length (Coffie, 1998; Underhill, 1949).

E. niger juveniles between the total lengths of 2 cm and 15 cm feed primarily on aquatic insect larva (Hunter & Rankin, 1939; Raney, 1942; Warner, 1973). Dragonfly and mayfly larva make up about 60% or more of *E. niger* diet at this stage of development (Hunter & Rankin, 1939; McLeod, 1903; Raney, 1942; Warner, 1973). The remainder of their diet is made up of other insect larva and small juvenile fish (Hunter & Rankin, 1939; Raney, 1942; Warner, 1973). *E. niger* undergo a distinct dietary shift as they continue to grow past 10 cm total length (Foote & Blake, 1945). During this shift, *E. niger* transition from relying mostly on aquatic insects nymphs to relying primarily on other fish as a source of food (Foote & Blake, 1945; Hunter & Rankin, 1939; Raney, 1942; Warner, 1973).

Growth is variable and depends on food availability, water chemistry, fish community associations and population density (Underhill, 1949). Once mature, average yearly growth is about 6.3 cm/year and 0.18 kg/year (Scott & Crossman, 1973). *E. niger* grow faster and reach sexual maturity faster in warmer environments. Sexual maturity may be reached within 1 year in warmer zones, compared to northern regions where it may take a *E. niger* 3 to 4 years to reach sexual maturity (Underhill, 1949). Females grow faster, mature earlier, attain a larger size and live longer than males (Coffie, 1998; Scott & Crossman, 1973). *E. niger* live an average of 3 to 4 years, but may attain an age of 8 to 9 years under ideal growing conditions; warm water temperatures and a plentiful supply of prey (Coffie, 1998; Scott & Crossman, 1973). The average size of adults ranges from about 38 to 46 cm with an average weight of 0.4 to 0.7 kg, maximum documented length is about 76 cm with a maximum weight of 2.7 kg (Scott & Crossman, 1973).

When fully mature, *E. niger* are primarily piscivorous on smaller fish as a (Coffie, 1998; Scott & Crossman, 1973). Mature chain pickerel will move into shallow waters to feed (Coffie, 1998). They spend more time in deeper water during the summer when temperatures are high (Coffie, 1998). Studies indicate that food selection for this species is only limited by gape size and body length; it can swallow prey where the body depth is less than or equal to its own body size (Coffie, 1998; Hunter & Rankin, 1939; Lee et al., 1980).

Many sources have documented the highly opportunistic predatory nature of mature *E. niger* (Coffie, 1998; Foote & Blake, 1945; Hunter & Rankin, 1939; Raney, 1942). In addition to its primary fish diet, *E. niger* has been documented to prey on frogs, tadpoles, snakes, baby turtles, salamanders, mice, leeches, and baby waterfowl (Coffie, 1998; Gilhen, 1999; Hunter & Rankin, 1939; Lee et al., 1980; MacLeod, 2020). However. Those types of opportunistic prey items make up a relatively small portion of *E. niger* diet compared to native fish species (Coffie, 1998; Hunter & Rankin, 1939; Lee et al., 1980).

1.3 Distribution

The native range *E. niger* range consists of the Atlantic Plain physiographic region from southwest Maine to southern Florida (Lee et al., 1980). Some native populations also exist from the Gulf Coast states west to the Sabine and Red river drainages in Texas, and within the Mississippi River basin north to Kentucky and Missouri (Hubbs et al., 1991; Lee et al., 1980). Populations are also listed as being native to eastern Texas (Hubbs et al., 1991; Lee et al., 1980). In Canada, native populations may only exist in the southwestern region of Quebec but there is some debate surrounding this claim (Hoyle & Lake, 2011; Page & Burr, 2010; Scott & Crossman, 1973).

E. niger have expanded outside their natural range via anthropogenic activities (Lee et al., 1980; Page & Burr, 2010). Many authors have attributed this to alterations in natural drainage for irrigation and land reclamation purposes (Crossman, 1991). Others note that *E. niger* have also been widely introduced illegally as a sport fish (Crossman, 1991; Livingstone, 1950; Page & Burr, 2010). In Atlantic Canada, *E. niger* are considered an introduced species to both New Brunswick and Nova Scotia (Scott & Crossman, 1959; Livingstone, 1950)

1.3.1 *Esox niger* in Nova Scotia

E. niger were introduced to Nova Scotia in the late 1940's and early 1950's (Gilhen & Pentz, 1974; Livingstone, 1950). There is no mention of *E. niger* in early fish surveys of the province prior to the 1940's, though this could be due to the limited number of surveyed lakes and survey effort (Gilhen, 1999). Records indicate that a sports fisherman introduced the species from the United States into the Spectacle Lake brook system near Comeauville in Digby county sometime in the 1920's (Gilhen & Pentz, 1974; Livingstone, 1950). The first documented occurrence of the species was on September 16, 1948 when a specimen was caught in Lac à Jeune (Young Lake), now Lac Innocent (Livingstone, 1950; Nova Scotia Department of Natural Resource, 1958). Several specimens were also collected at Upper and Lower Spectacle Lakes (now Spectacle Lake and Lac d'en Bas or Lower Lake respectively) between July 26, 1949 and July 30, 1949 (Gilhen, 1969; Gilhen & Pentz, 1974; Livingstone, 1950).

Since their introduction, *E. niger* have spread throughout Nova Scotia. In 1986, *E. niger* were reported within nine surveyed lakes in Yarmouth county (Alexander et al., 1986). By 2010, *E. niger* were documented in over 95 known locations, as far as Blacketts and Gillis Lakes in Cape Breton (Cape Breton Post, 2012; Mitchell et al., 2011; Swinemar et al., 2021). By 2017, *E.*

niger were documented in 112 distinct water bodies, and by 2019, this number increased to 136 and by 2022, 201 distinct waterbodies (Mitchell et al., 2011; Swinemar et al., 2021). *E. niger* now occurs in twelve out of the eighteen counties in Nova Scoti, including Yarmouth, Shelburne, Digby, Lunenburg, Kings, Hants, Halifax, Queens, Annapolis, Colchester, Pictou, and Cape Breton counties (Swinemar et al., 2021) (Figure 4).

1.3.2 *Esox niger* in Kejimkujik National Park and National Historic Site

The first report of *E. niger* within Kejimkujik National Park and National Historical Site was on June 24, 2018, in a section of the Mersey River called "The Dump" near the end of portage "O" (Parks Canada, 2019; Swinemar et al., 2021) (Table 1). *E. niger* entered KNPNHS via the Mersey River (Parks Canada, 2019; D. Reid, personal communication, January 21, 2020). The fish likely entered the Mersey via Lake Rossignol from outside the park. *E. niger* were first documented in Lake Rossignol near the mouth of the Shelburne River in April of 2018 (D. Reid, personal communication, January 21, 2020; Swinemar et al., 2021). The thinking is that this fish species arrived from the Jordan River system from Jordan Lake in 1995 via an Nova Scotia Power artificial canal connecting Jordan Lake to Silver Lake Brook (MacEachern, 1956; Nova Scotia Power, 2018; D. Reid, personal communication, January 21, 2020; Swinemar et al., 2021). Silver Lake Brook flows from Silver Lake into Sixth Lake, Sixth Lake is connected to Lake Rossignol via the Sixth Lake Brook (MacEachern, 1956; Nova Scotia Power, 2018; D. Reid, personal communication, January 21, 2020; Swinemar et al., 2021).

By 2020, *E. niger* have been confirmed in seven KNPNHS lakes (Loon, George, Kejimkujik, Grafton, Peskowesk, Frozen Ocean, Big Dam West Lake, and Cobrielle) (Parks Canada, 2019; Swinemar et al., 2021). Just outside of the KNPNHS boundaries, *E. niger* have

been confirmed in connecting West River, Rogers Brook, and the Mersey River up until the first set of falls at Mill Falls (Parks Canada, 2019; Swinemar et al., 2021). Further consideration must be given to Beaverskin, Pebbleloggitch, and Peskawa lakes, all of which are at risk of invasion due to connectivity with already invaded lakes.

1.3.3 *Esox niger* in New Brunswick

No species of *Esox* pike or pickerel were present in New Brunswick before the year 1850 (Perley, 1850). The first official observation of *E. niger* was in October 23, 1893, reported by Dr. William Kendall who noted 12 cm – 13 cm fish in the New Brunswick side of St. Croix River near Baring (Scott & Crossman, 1959; United States Fish Commission, 1894).. This coincided with the artificial introduction of *E. niger* into the Grand Lake (Maine) portion of the St. Croix River system in 1863 (Adams, 1873). Shortly after Kendall made his observations, *E. niger* were introduced into the St. John river system via the Meduxnakik River (Cox, 1899).

At present, *E. niger* are abundant within both the St. Croix and St. John River Basins (Canadian Rivers Institute, n.d.; Swinemar et al., 2021). *E. niger* now occurs in 143 distinct water bodies across ten out of the fifteen counties in New Brunswick,including Victoria, Carleton, York, Queens, Sunbury, Kings, Saint John, Charlotte, Albert, and Westmorland counties (Figure 5).

1.4 The Impacts of Invasive *Esox niger* in Nova Scotia and Knowledge Gaps

Many anglers and researchers have expressed concerns over the impacts of invasive *E. niger* on the native fish populations in Nova Scotia's lakes and streams (Alexander et al., 1986; Crowley, 2018; Livingstone, 1950; Mitchell et al., 2011).

E. niger are considered to be extremely inimical towards native fresh water fish species within their non-native range (Alexander et al., 1986; Gilhen & Pentz, 1974; Livingstone, 1950). They have been cited as negatively effecting trout, salmon, cyprinid, and other piscivorous fish communities, as well as other native *Esox* species across their non-native range (Alexander et al., 1986; Gilhen & Pentz, 1974; Hoyle & Lake, 2011; Livingstone, 1950).

Overall *E. niger* results in the replacement of a traditionally highly valued recreational fishery with one of lesser value (Mitchell et al., 2011). It was found that fish species richness and diversity is higher in non-invaded lakes than in *E. niger* invaded lakes (Mitchell et al., 2011). There is a total loss of small-bodied fish species and a truncation of fish body size distribution, leaving only larger native fish species (Mitchell et al., 2011). The presence of *E. niger* is also likely to result in changes in lake functioning and may possibly alter the complexity of the trophic food web (Mitchell et al., 2011).

A study by MacLeod 2020 investigated the diet of invasive *E. niger* and *Micropterus dolomieu* (Smallmouth Bass) in the LaHave River system (MacLeod, 2020). A non-lethal gastric lavage technique was used to remove stomach contents, which were analyzed using gravimetric and volumetric measurements (MacLeod, 2020). It was shown that invertebrates occurred most frequently in 88% of *E. niger* samples. The Order Odonata, Anisoptera and Zygoptera, were the most consumed invertebrates, making up 89.7% of invertebrates consumed by *E. niger* (MacLeod, 2020). Fish composed the greatest proportion for *E. niger* stomach contents, consisting of 76% of the total wet weight of stomach contents. The data in this study suggested that there is potential that Chain Pickerel are negatively affecting native biota within Nova Scotia.

A recent honors thesis by Brake in 2020 investigated the response of native fish

populations to introduced *E. niger* in KNPNHS (Brake, 2020). Alaska trap nets were set at select study lakes where fish were monitored. *E. niger* were found to be the largest, or nearly so, of all fish species in KNPNHS (Brake, 2020). *E. niger* are likely the reason for declines in abundances of many fish species, and a decrease in abundances of smaller individual fish (Brake, 2020).

Introduced *E. niger* are strongly associated with the overall decline of native fish species within invaded systems and alteration of food web structures (Brake, 2020; MacLeod, 2020; Mitchell et al., 2011). Based on the information presented in this section, several knowledge gaps have been identified:

1. Are *E. niger* feeding preferably on any specific carbon sources such as benthic invertebrates, native fish or pelagic sources?
2. Are *E. niger* dietary preferences related with their size class?
3. How are *E. niger* affecting food web structure over time?

1.5 An overview of Stable Isotope Analysis

Over the last 3 decades, stable isotope analysis (SIA) has been used to assess structure and dynamics of aquatic food webs (Layman et al., 2007; Post, 2002). Data modelling methodologies now range from qualitative inferences based on the isotopic niche, to Bayesian mixing models that can be used to characterize food-web structure and dietary niches (Jackson et al., 2011; Layman et al., 2007). SIA data are expressed as delta (δ) values in per mil (‰). The equation below is used to calculate δ values (Fry, 1988):

$$\delta X = \left[\left(\frac{R_{sample}}{R_{std}} - 1\right)\right] \times 1{,}000 \quad (1)$$

Where δX represents the isotope of interest and "*R*" represents the ratio of the isotope of interest and its natural form (a standard) (Fry, 1988).

Stable isotope ratios of carbon-13/carbon-12 and nitrogen-15/nitrogen-14 are most used in food web analysis and will be the focus moving forward. Several other isotopes such as δ-sulfur-34, δ-oxygen-18, and δ-hydrogen-2 can be used to examine different aspects of natural systems (Fry, 2008; Michener et al., 2007). Carbon stable isotopes (carbon-13 and carbon-12) can be used to determine the primary production source responsible for the energy flow in an aquatic ecosystem (Layman et al., 2007; Post, 2002). Plants contain less ^{13}C than atmospheric CO_2 used for photosynthesis (Michener et al., 2007). The plants are therefore lower ^{13}C relative to the atmospheric concentrations. The lower ratio of 13C relative to 12C is due to enzymatic and physical processes that discriminate against ^{13}C in favor of ^{12}C (Michener et al., 2007). In fish and other aquatic animals, decarboxylation is responsible for this process. Discrimination varies among plants using different photosynthetic pathways (Michener et al., 2007). The Calvin cycle (C3), Hatch-Slack cycle (C4) and Crassulacean acid metabolism (CAM) photosynthetic pathways differ profoundly and consistently enough that $\delta^{13}C$ isotopic signatures can be used to distinguish between them (Michener et al., 2007). The transfer of ^{13}C throughout trophic levels remains relatively consistent, only having a small increase (trophic fractionation $\sim< 1$ ‰) per trophic level (Post, 2002).

Nitrogen stable isotope ratios (nitrogen-15 and nitrogen-14) can provide an indication of an organism's trophic position (Post, 2002). The $\delta^{15}N$ value of animal tissues is often used to indicate trophic position within aquatic food webs (Michener et al., 2007). Plants may take up either NO_3-, NH_4+, or dissolved organic nitrogen (DON), with many (but not all) plants showing distinct preferences (Michener et al., 2007). The loss of nitrogen isotopes through deamination is

accompanied by significant isotopic fractionation and appears to be the primary factor contributing to the trophic fractionation of an organisms tissues relative to its food source (trophic fractionation ~3.4‰) (Michener et al., 2007). As organisms eat each other, ^{14}N is preferably lost through urine and excretion, while ^{15}N are transferred to the predators (Layman et al., 2007; Post, 2002). Consequently, organisms higher in the trophic pyramid will have accumulated higher levels of ^{15}N to ^{14}N (e.g., higher $\delta^{15}N$ values) relative to their prey and in the food web (Layman et al., 2007; Post, 2002).

The $\delta^{15}N$ and $\delta^{13}C$ values of various organisms are commonly plotted together to form a bi-plot (Bodey et al., 2012; Layman et al., 2007) (Figure 6), aka δ-space or bi-plot space (Layman et al., 2007; Post, 2002). The relative position of species to each other in a bi-plot space is used to infer aspects of food web structure (Layman et al., 2007; Post, 2002). Advancements in stable isotope models such as MixSIAR have made it possible to approximate the relative tropic position and dietary source proportions for various units in a food web analyses using stable isotope data (Jackson et al., 2011; Layman et al., 2007; Post, 2002).

1.5.1 Layman Metrics

Layman metrics are a set of 6 metrics commonly used to examine relative areas and positions of the centroid of each species clusters within food web structure biplots (Layman et al., 2007). The first four of these metrics are used to measures the spacing of isotope values in δ-space (community-wide measures of trophic diversity) (Layman et al., 2007). The final two metrics reflect relative position of species to each other within niche space and can be used to estimate the extent of trophic density (Layman et al., 2007). These metrics are defined below by Layman as:

- $\delta^{15}N$ Range (NR): Distance between the two species with the lowest ^{15}N and highest $\delta^{15}N$ values (i.e., maximum $\delta^{15}N$ - minimum $\delta^{15}N$) (Layman et al., 2007). NR is one representation of vertical structure within a food web (Layman et al., 2007).
- $\delta^{13}C$ range (CR): Distance between the two species with the lowest ^{13}C and highest $\delta^{13}C$ values (i.e., maximum $\delta^{13}C$ - minimum δ13C) (Layman et al., 2007). Increased CR would be expected in food webs in which there are multiple basal resources with varying $\delta^{13}C$ values (Layman et al., 2007)
- Total area (TA): Convex hull area encompassed by all species in $\delta^{15}N$ - $\delta^{13}C$ bi-plot space (Layman et al., 2007). This represents a measure of the total amount of niche space occupied, and thus a proxy for the total extent of trophic diversity within a food web (Layman et al., 2007).
- Mean distance to centroid (CD): Average euclidean distance of each species to the $\delta^{15}N$ - $\delta^{13}C$ centroid, where the centroid is the mean $\delta^{13}C$ and $\delta^{15}N$ value for all species in the food web (Layman et al., 2007). This metric provides a measure of the average degree of trophic diversity within a food web (Layman et al., 2007).
- Mean nearest neighbour distance (NND): Mean of the Euclidean distances to each species nearest neighbor in bi-plot space, and thus a measure of the overall density of species packing (Layman et al., 2007).
- Standard deviation of nearest neighbour distance (SDNND): A measure of the evenness of species packing in bi-plot space that is less influenced than NND by sample size (Layman et al., 2007).

1.5.2 Lipid Correction

Natural variations exist in stable isotope ratios within various tissue types such as lipids, protein and carbohydrates (Gannes et al., 1997; Kiljunen et al., 2006; Sotiropoulos et al., 2004). Lipids

tend to have lower ^{13}C relative to ^{12}C in comparison to other tissue components (DeNiro & Epstein, 1978; Kiljunen et al., 2006; Thompson et al., 2000). High lipid content of sampled muscle tissues can complicate the interpretation of dietary carbon because the average tissue $\delta^{13}C$ can be highly impacted by high lipid content in a tissue sample which adds extra variability and bias in data interpretation when comparing muscle $\delta^{13}C$ values between multiple species (Focken & Becker, 1998; Rolff & Elmgren, 2000). Lipid-corrected $\delta^{13}C$ values are more indicative of assimilated carbon in muscle tissue, while uncorrected $\delta^{13}C$ values reflect the combined effects of assimilation and lipid synthesis processes (Power et al., 2003).

Lipid correction is a process used to address the influence of lipids on stable isotope values, particularly $\delta^{13}C$ (Kiljunen et al., 2006; Saini et al., 2021). Lipids have a comparatively lower carbon isotope ratio compared to proteins and carbohydrates (Kiljunen et al., 2006; Saini et al., 2021). Lipid correction is critical in stable isotope studies where $\delta^{13}C$ values are used to infer dietary sources and trophic positions (Kiljunen et al., 2006; Saini et al., 2021). A common approach involves the chemical extraction of lipids from tissues, followed by re-analysis of the lipid-free tissues for stable isotope composition (Sotiropoulos et al., 2004). Over time, predictive models for lipid correction have been developed based on extensive datasets comparing stable isotope values of samples before and after lipid extraction (Kiljunen et al., 2006). These models often use tissue C ratios as proxies to estimate lipid content and provide corrections without the need for chemical extraction (Saini et al., 2021).

One of the most used non-chemical lipid normalizing models is a refined version (Kiljunen et al., 2006) of two normalization models developed by McConnaughey and McRoy (1979) and Alexander et al. (1996) respectively (Alexander et al., 1996; McConnaughey & McRoy, 1979). This model is as follows:

$$\delta^{13}C_{lipid} = \delta^{13}C + D \cdot \left(I + \frac{3.90}{1 + 287/L}\right) \quad (2)$$

Where $\delta^{13}C_{lipid}$ is the lipid-corrected value. The first parameter, D, is the isotopic difference between protein and lipids and defines the slope curvature of the model and is defined as D = 7.018 ± 0.263 (Kiljunen et al., 2006). The second parameter, I, defines the intersection on the x-axis and is defined as I = 0.048 (Kiljunen et al., 2006). Lastly, L, is the proportional lipid content of the sample and is defined by:

$$L = \frac{93}{1 + (0.246 \cdot (C:N) - 0.775)^{-1}} \quad (3)$$

Where C : N is the proportion of total carbon and total nitrogen in the sample.

1.5.3 Cluster Analysis

Cluster analysis is a common statistical analysis method used to organize data into groups or clusters (Santos et al., 2018). Cluster analysis can be used to group consumers or sources based on $\delta^{13}C$ and $\delta^{15}N$ values in δ-space. *E. niger* feed on benthic macroinvertebrates (BMI) and native fish sources preferentially depending on their total length (Brake, 2020; Hunter & Rankin, 1939; MacLeod, 2020; Raney, 1942; Warner, 1973). Odonata nymphs and native fish group means usually appear at distinctly different $\delta^{15}N$ values (Clayden et al., 2013; Ofukany et al., 2014; Vander Zanden et al., 1999; Zanden & Rasmussen, 2001). Assuming this is the case, *E. niger* may be grouped into two distinct clusters in δ-space based on their preferred source. These

two *E. niger* clusters will be referred to as CP1 and CP2 in this section. This assumption will later be verified in chapter 2.

The NbClust is an R package commonly used to perform cluster analysis (Charrad et al., 2014). NbClust can be used to assign individual *E. niger* into optimal non overlapping clusters based on their $\delta^{15}N$ and $\delta^{13}C$ stable isotope values (Charrad et al., 2014). This provides insight into their trophic position and dietary sources. A new column is typically added to the dataset to indicate the cluster assignment, in this case for every *E. niger*. Individuals in the first cluster ($4.1‰ \leq \delta^{15}N \leq 6.4‰$, $-30.1‰ \leq \delta^{13}C \leq -28.9‰$) are labeled as CP1, representing those with lower $\delta^{15}N$ and $\delta^{13}C$ values, possibly reflecting reliance on specific food sources. Those in the second cluster ($7.1‰ \leq \delta^{15}N \leq 10.4‰$, $-30.6‰ \leq \delta^{13}C \leq -25.7‰$) are labeled as CP2, indicative of higher $\delta^{15}N$ and $\delta^{13}C$ values, suggesting a different dietary pattern or trophic position.

There exists a centroid point in $\delta -$ space, denoted here as $\Psi(\delta^{13}C, \delta^{15}N)$, between clusters CP1 and CP2 (Bandyopadhyay & Saha, 2012; Everitt et al., 2001). The centroid point Ψ is the transition point between groups CP1 and CP2 and is analogous to *E. niger* dietary transition. Ψ can be calculated by utilizing average linkage clustering; first by finding the midpoint of each pairwise distance and then averaging all pairwise midpoints in $\delta -$ space using equations (*4*) and (*5*) (Bandyopadhyay & Saha, 2012; Everitt et al., 2001):

$$\Psi(\delta^{13}C, \delta^{15}N) = \left(d_{ij}(\delta^{13}C), d_{ij}(\delta^{15}N)\right) \tag{4}$$

$$d_{ij} = \frac{1}{2kl}\sum_{i=1}^{k}\sum_{j=1}^{l} D\left(\boldsymbol{A_i}, \boldsymbol{B_j}\right) \tag{5}$$

Where d_{ij} is the centroid between two clusters, *k* and *l* are the number of points belonging to clusters one and two (CP1 and CP2) respectively (Bandyopadhyay & Saha, 2012). $\boldsymbol{A_1}, \boldsymbol{A_2}, \cdots \boldsymbol{A_k}$ and $\boldsymbol{B_1}, \boldsymbol{B}, \cdots \boldsymbol{B_l}$ are observations from clusters one and two respectively. $D(\boldsymbol{A_i}, \boldsymbol{B_j})$ is the pairwise distance between two points belonging to $\boldsymbol{A_k}$ and $\boldsymbol{B_l}$ (Bandyopadhyay & Saha, 2012; Everitt et al., 2001). The centroid point in delta space between two clusters is defined by Equation (*6*):

$$\Psi = \left(\frac{1}{2kl} \sum_{i=1}^{k} \sum_{j=1}^{l} D\left(A_i(\delta^{13}C), B_j(\delta^{13}C)\right) \ , \frac{1}{2kl} \sum_{i=1}^{k} \sum_{j=1}^{l} D\left(A_i(\delta^{15}N), B_j(\delta^{15}N)\right) \right) \quad (6)$$

The uncertainty associated with Ψ is determined using:

$$\sigma_{ij} = \frac{1}{kl} \sum_{i=1}^{k} \sqrt{\sum_{j=1}^{l} \frac{\left(D\left(\boldsymbol{A_i}, \boldsymbol{B_j}\right) - \mu\right)^2}{2}} \quad (7)$$

Where μ in the mean and is defined as:

$$\mu = \frac{\sum_{j=1}^{l} D\left(\boldsymbol{A_i}, \boldsymbol{B_j}\right)}{l} \quad (8)$$

Average linkage clustering can be applied to total length data. There exists a centroid point, denoted here as Ω(*TL*), between clusters CP1 and CP2 (Bandyopadhyay & Saha, 2012; Everitt et al., 2001). The centroid point Ω is significant, it represents the total length associated with the transition point between groups CP1 and CP2. Ω can be calculated utilizing average linkage clustering; first by finding the midpoint of each pairwise distance and then averaging all pairwise midpoints (Bandyopadhyay & Saha, 2012; Everitt et al., 2001). In this case we are dealing with one dimension rather than two as was the case in δ-space:

$$\Omega(TL) = \left(d_{ij}(TL)\right) \tag{9}$$

$$d_{ij} = \frac{1}{2kl}\sum_{i=1}^{k}\sum_{j=1}^{l} D\left(\boldsymbol{A_i}, \boldsymbol{B_j}\right) \tag{10}$$

Where d_{ij} is the centroid between two clusters, *k* and *l* are the number of points belonging to clusters one and two (CP1 and CP2) respectively (Bandyopadhyay & Saha, 2012; Everitt et al., 2001). $\boldsymbol{A_1}, \boldsymbol{A_2}, \cdots \boldsymbol{A_k}$ and $\boldsymbol{B_1}, \boldsymbol{B}, \cdots \boldsymbol{B_l}$ are observations from clusters one and two respectively. $D(\boldsymbol{A_i}, \boldsymbol{B_j})$ is the pairwise distance between two points belonging to $\boldsymbol{A_k}$ and $\boldsymbol{B_l}$ (Bandyopadhyay & Saha, 2012; Everitt et al., 2001). The centroid point in delta space between two clusters is defined by Equation (11):

$$\Omega(TL) = \left(\frac{1}{2kl}\sum_{i=1}^{k}\sum_{j=1}^{l} D\left(A_i(TL), B_j(TL)\right)\right) \tag{11}$$

The uncertainty associated with Ω is determined using:

$$\sigma_{ij} = \frac{1}{kl}\sum_{i=1}^{k}\sqrt{\sum_{j=1}^{l}\frac{\left(D\left(\boldsymbol{A_i}, \boldsymbol{B_j}\right) - \mu\right)^2}{2}} \quad (12)$$

Where μ in the mean and is defined as:

$$\sigma_{ij} = \frac{1}{kl}\sum_{i=1}^{k}\sqrt{\sum_{j=1}^{l}\frac{\left(D\left(\boldsymbol{A_i}, \boldsymbol{B_j}\right) - \mu\right)^2}{2}} \quad (13)$$

1.5.3.1 Optimal Number of Clusters

The optimal number of clusters to assign can be determined using the R NbClust package, which offers a comprehensive suite of 30 different indices to evaluate clustering performance (Charrad et al., 2014). Each of the 30 indices calculates the optimal numbers of clusters (k), reflecting various aspects of clustering performance such as compactness and separation (Charrad et al., 2014). For instance, the Calinski and Harabasz Index (CH) evaluates the ratio of between-cluster dispersion to within-cluster dispersion, while the Silhouette Index measures how similar an object is to its own cluster compared to others (Charrad et al., 2014). NbClust identifies the optimal k for each of the 30 indices based on their respective criteria (Charrad et al., 2014). The NbClust package then aggregates these results, using a majority voting approach to determine the most frequently recommended number of clusters (Charrad et al., 2014). By applying these indices, we were able to obtain a robust and reliable determination of the optimal number of clusters, ensuring the validity and reliability of our subsequent analyses

(Charrad et al., 2014). The use of multiple indices provided a well-rounded evaluation, as each index has its own strengths and limitations (Charrad et al., 2014).

1.5.4 Mixing Models

Mixing models are analytical tools used in stable isotope analysis to estimate the proportional contributions of multiple sources to a mixture (Bond & Diamond, 2011; Fry, 2013). The mixture consists of your consumer of interest and their suspected sources. Mixing models use the isotopic signatures (ratios of stable isotopes, such as $\delta^{13}C$ and $\delta^{15}N$) of the sources and the mixture to estimate the proportions of each source in the mixture (Bond & Diamond, 2011; Fry, 2013).

Unlike gut content analysis, which provides a snapshot of recent feeding, stable isotope mixing models provide an integrated view of diet over a longer period (Parzanini et al., 2019). This is possible because stable isotopes in animal tissues have specific turnover rates, which are the times it takes for the isotopic signature in a tissue to reflect changes in diet or environment (Jardine et al., 2003; Parzanini et al., 2019). Different tissues have different turnover rates, allowing researchers to study dietary habits over various timescales (Jardine et al., 2003; Parzanini et al., 2019). For instance, blood might have a turnover rate of weeks, reflecting recent dietary changes, while bone might have a turnover rate of months or years, indicating long-term dietary patterns (Jardine et al., 2003; Parzanini et al., 2019). These turnover rates are crucial for interpreting stable isotope data accurately, as they help determine the timeframe over which dietary information is integrated, offering insights into both short-term and long-term feeding behaviors (Jardine et al., 2003; Parzanini et al., 2019).

MixSIAR (Mixing Models for Stable Isotope Analysis in R) is a popular mixing model used to analyze stable isotope data. By utilizing Bayesian mixing models, MixSIAR estimates the proportions of various sources in the diet of the consumers (Stock & Semmens, 2016). Bayesian statistics allow MixSIAR to handle multisource situation (Stock & Semmens, 2016). Bayesian mixing models provide probability distributions for the estimated source contributions rather than single point estimates (Stock & Semmens, 2016).

MixSIAR and other Bayesian mixing models can incorporate prior knowledge about the source contributions, this can help refine estimates, especially when data are limited (Stock & Semmens, 2016). MixSIAR utilizes informative or uninformative priors often referred to as alpha (α) priors. Gut content data is commonly used to form alpha priors. Gut content reflects the actual diet of a consumer and therefore acts as a good informative prior for influencing the model.

1.6 Stable Isotope Analysis and Invasive Ecology

Stable isotope analysis can be used to determine the effects of an introduced species on native aquatic food web structure (Bodey et al., 2012). Invasive species often differ functionally from the organisms of a recipient community (Gallardo et al., 2016). Because of this, an invasive species will generate ecological impacts that propagate along native food web structure (Gallardo et al., 2016). The ecological impacts of invasive species are reflected within changing stable isotope values (Bodey et al., 2012; Layman et al., 2007). The dynamic characteristics of stable isotope values can be quantified either temporally or spatially (Layman et al., 2007; Michener et al., 2007). Temporal comparisons of organism stable isotope values can be made at different stages of invasion to determine the impacts of an invasive species over time, with each sample

set collected at specific times representing a temporal snapshot of the food web (Gallardo et al., 2016; Layman et al., 2007; Michener et al., 2007). Spatial comparisons of isotope values can be made by comparing similar systems at different stages of invasion to determine the impacts of an invasive species (Gallardo et al., 2016; Layman et al., 2007; Michener et al., 2007).

SIA can be used to determine the effects invasive species have on aquatic food webs. It is important to consider the impacts of invasive species as context-dependent, differing between species and habitats (Bodey et al., 2012; Gallardo et al., 2016). Invaders that differ functionally from native species can have varying effects on food web structure. In some cases, impacts propagate up and down food webs, as in the case of species that are in filtering, collecting and predator niches (Gallardo et al., 2016). In others, changes dissipate within one functional level, suggesting compensatory effects to the introduction of invasive species, such as the presence of refuges, the ability to shift food sources (in the case of omnivores) and mechanisms to avoid predation (Gallardo et al., 2016). In the case of the introduction of a new top predator species, this change would be reflected through Leyman metrics by an increase in NR and TA (Gallardo et al., 2016; Layman et al., 2007).

1.7 Stable Isotopes and the Impacts of *Esox* species on Native Freshwater Food Web Structure

Stable isotope analysis can be used to investigate the impacts invasive species have on freshwater ecosystems. In the preceding sections we explored SIA and how it may be applied to understand how the introduction of invasive species can effect the original native aquatic food web structure. The effects *Esox* species have on food web structure can be determined effective by comparing pre-pickerel SIA data with new post-pickerel SIA data, either spatially or

temporally. There are several ways of understanding this; we can apply local knowledge of *E. niger* to better understand their impact, or we can examine similar studies completed using different *Esox* species.

This section will combine knowledge gained in previous section with the results of SIA studies that focus on defining the role of *Esox* species. Specifically, the Northern pike, *E. lucius,* is much more widely studied and assessed in food webs globally, this species will be included in a literature review here to develop theories about the possible impacts of *E. niger* and the effects it may have on Nova Scotia's freshwater ecosystems. We are able to make these comparisons because *Esox* species consistently exhibit similar behaviour across species (Coffie, 1998; Lee et al., 1980; Scott & Crossman, 1973).

Similarly to *E. niger*, *E. lucius* is considered to be piscivorous throughout most of its range (Venturelli & Tonn, 2006). Its morphology and behaviour are specialized for ambushing fish prey from the cover of vegetation (Venturelli & Tonn, 2006). Unlike *E. niger,* there have been numerous studies completed that investigate the food web dynamics of *E. lucius*, some of which utilize SIA. Several papers have been published that investigate the trophic adaptability of *E. lucius* both as native and as an invasive species (Beaudoin et al., 1999; Venturelli & Tonn, 2006).

In Northern Alberta, stable isotope analysis (SIA) tools were used to examine the trophic adaptability and dietary flexibility of native *Esox lucius* (Northern Pike) in relation to varying food web structures (Beaudoin et al., 1999). This study focused on comparing the trophic ecology of pike across several lakes with distinct ecological scenarios: *pike-only lakes* and *pike-other lakes*. In pike-only lakes, *E. lucius* is the sole fish species, leading to a food web dominated by invertebrate prey and demonstrating pike's ability to occupy lower trophic positions. In

contrast, pike-other lakes contain *E. lucius* alongside a variety of other native fish species, where pike assume a higher trophic position by preying on other fish species. This spatial comparison provided insights into how *E. lucius* adjusts its feeding strategy and trophic position based on the availability of prey resources and the complexity of the food web (Beaudoin et al., 1999).

By comparing SIA data with stomach content analysis (SCA) *E. lucius* were demonstrated to have excellent trophic adaptability (Beaudoin et al., 1999). *E. lucius* in pike-only lakes have lowerd ^{15}N in comparison to pike-other lakes, this is reflected in the δ15N axis (Beaudoin et al., 1999). *Esox lucius* are capable of shifting their trophic position to feed on lower or higher-level prey items (Beaudoin et al., 1999).

A second study after this one looked at (Beaudoin et al, 1999), 3 shallow fishless lakes with introduced *E. lucius* and compared to 2 lakes which already had Northern Pike regarding the trophic adaptability of *E. lucius* (Venturelli & Tonn, 2006). Introduced *E. lucius* were monitored for diet and growth over two summers (Venturelli & Tonn, 2006). Stomach content analysis revealed that stocked adults responded to the sudden absence of prey fishes by specializing on energy-rich leeches, whereas juvenile consumed a broader mix of invertebrates (Venturelli & Tonn, 2006).

Invasive *E. lucius* can have detrimental effects on native fish species (Haught & von Hippel, 2011). Sometime during the 1950's, *E. lucius* were introduced to the northern Susitna Basin of south central Alaska (Haught & von Hippel, 2011). Since their introduction *E. lucius* have spread throughout the upper Cook Inlet Basin (Haught & von Hippel, 2011). Extirpations of several native fish populations have been documented in this area (Haught & von Hippel, 2011). It is hypothesized here that invasive pike remodel the ecology of lakes by removing vulnerable prey types (Haught & von Hippel, 2011). The Alaska study assessed and compared

the diets of several native fish species with *E. lucius* using a relative importance index (Haught & von Hippel, 2011).The results of this study suggest that invasive *E. lucius* have a consistently detrimental effect on the continued existence of native fish populations because of their high trophic adaptability (Haught & von Hippel, 2011). Like previous studies discussed, the Alaska *E. lucius* can be sustained by a variety of prey sources (Beaudoin et al., 1999; Haught & von Hippel, 2011; Venturelli & Tonn, 2006). This allows them to supplement their diet with less desirable prey as preferred prey are reduced in abundance; in the case of this study native fish are supplemented with macroinvertebrates (Haught & von Hippel, 2011). *Esox lucius* are consequently able to thrive and apply predation pressure on native fish regardless of native fish abundance (Haught & von Hippel, 2011).This has ultimately resulted in native fish population declines and extirpations with in the Cook Inlet Basin and other part of Alaska where *E. lucius* are considered invasive (Haught & von Hippel, 2011).

The degree to which populations of native fishes are reduced likely depends on characteristics of the habitat in which the invasion occurs (Haught & von Hippel, 2011). Despite the significance lake characteristics play in the severity of species reduction or total extirpation, introduction of pike has been shown to have a negative effect on native fish abundance regardless of lake type (Haught & von Hippel, 2011).In the case of the Cook Inlet Basin, *E. lucius* have resulted in the loss of loss of native anadromous fishes (Haught & von Hippel, 2011).This has resulted in effects that are far reaching, as the delivery of marine derived nutrients to oligotrophic systems is being halted (Haught & von Hippel, 2011).

The introduction of a non-native top predator such as *Esox* spp. will cause a subsequent reduction and loss of native fishes (Gallardo et al., 2016; Haught & von Hippel, 2011). This will likely result in food web cascading effects where the structure and functioning of aquatic

communities are ultimately simplified (Gallardo et al., 2016; Haught & von Hippel, 2011). The cascading effect occurs when a *Esox* spp. is introduced into a new system, this radically alters predation regimes; gaining a new top predator results in the reduction of native fish and their previous predators (Gallardo et al., 2016; Haught & von Hippel, 2011). The introduction of a new top predator will also alter competitive regimes; for instance, some macroinvertebrates will be spared while others will be targeted (Gallardo et al., 2016; Haught & von Hippel, 2011; Venturelli & Tonn, 2006).

1.8 Summary

Like *E. lucius, E. niger* are a sight-oriented piscivorous fish species. They are considered an invasive species in Nova Scotia where they were introduced in the 1920's. As of 2022, *E. niger* have spread to 201 distinct freshwater systems within Nova Scotia. They were first detected Kejimkujik National Park and National Historic Site (KNPNHS) in 2018. KNPNHS was protected to preserve a representative portion of the Atlantic upslope region that harbours a unique ecosystem defined by unique plant and wildlife species. The effects of *E. niger* on native freshwater species and overall food web structure are not well understood. Based on invasive *Esox* literature elsewhere, many authors hypothesize that *E. niger* modify food web structure by removing preferred prey types resulting in an overall loss of biodiversity. This thesis will focus on addressing several key questions aimed at broadening our understanding of the impacts of *E. niger* in KNPNHS:

1. Are *E. niger* feeding preferably on any particular carbon source such as benthic invertebrates, native fish or pelagic zooplankton sources?
2. Is *E. niger* diet related to size class?
3. How are *E. niger* affecting food web structure over time?

1.9 Figures

Figure 1: An E. niger caught in the Mersey River within Kejimkujik National Park and National Historic Site (KNPNHS).

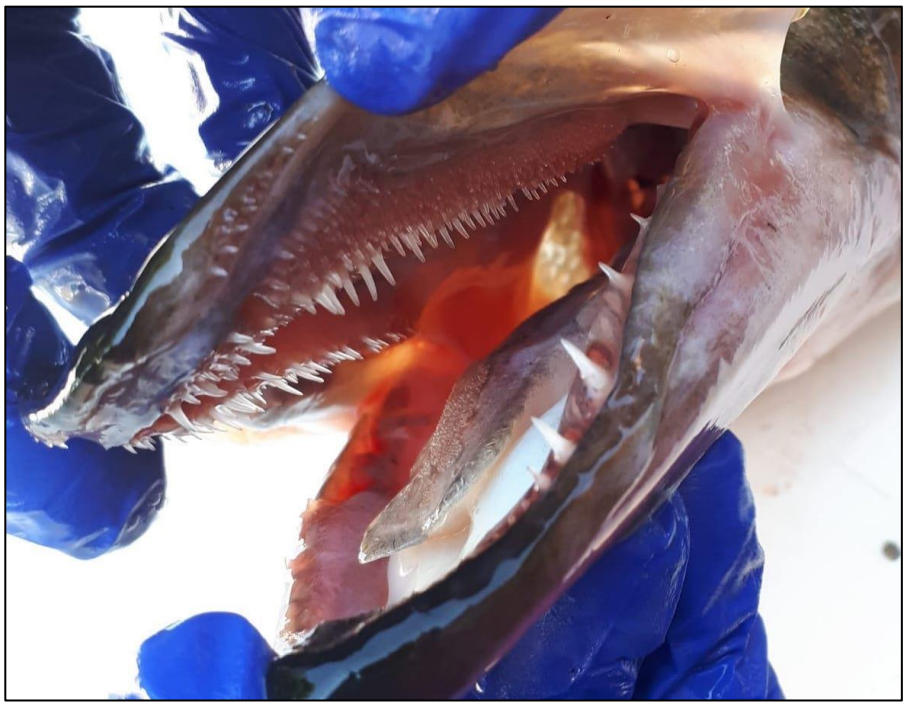

Figure 2: E. niger have several rows of teeth on their upper jaw and one row on their lower jaw

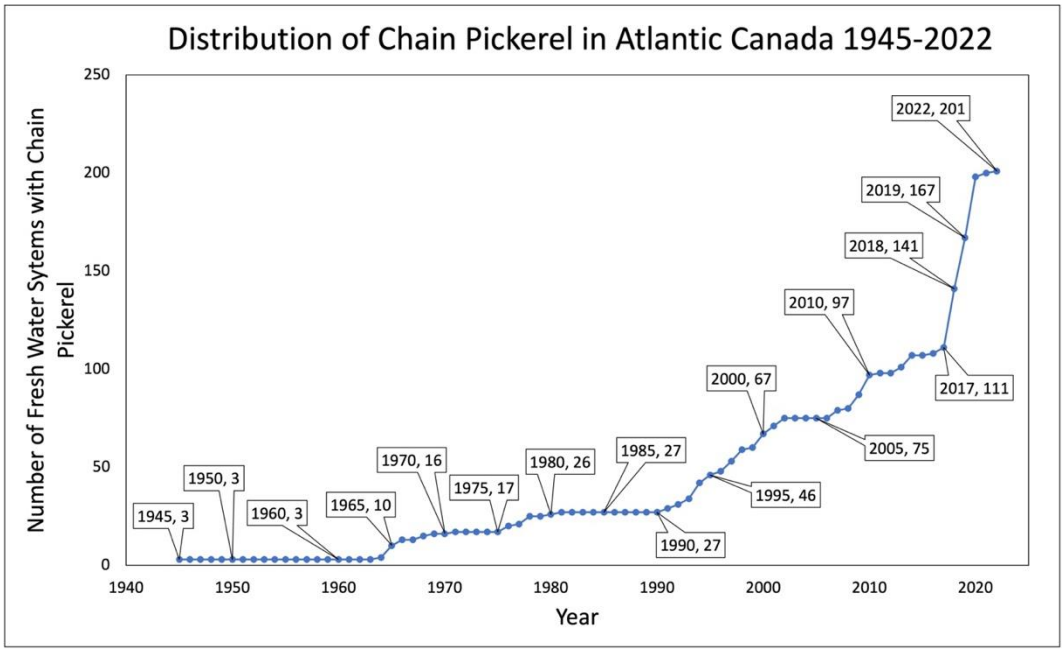

Figure 3: Cumulative distribution of number of known lakes in Nova Scotia documented to contain E. niger each year, 1945-2022. Total number of distinct water bodies known to contain pickerel in 2022 = 201. Data provided by the Atlantic Canada Chain Pickerel Compiled Database and Map (Swinemar et al., 2021).

Nova Scotia Chain Pickerel Distribution

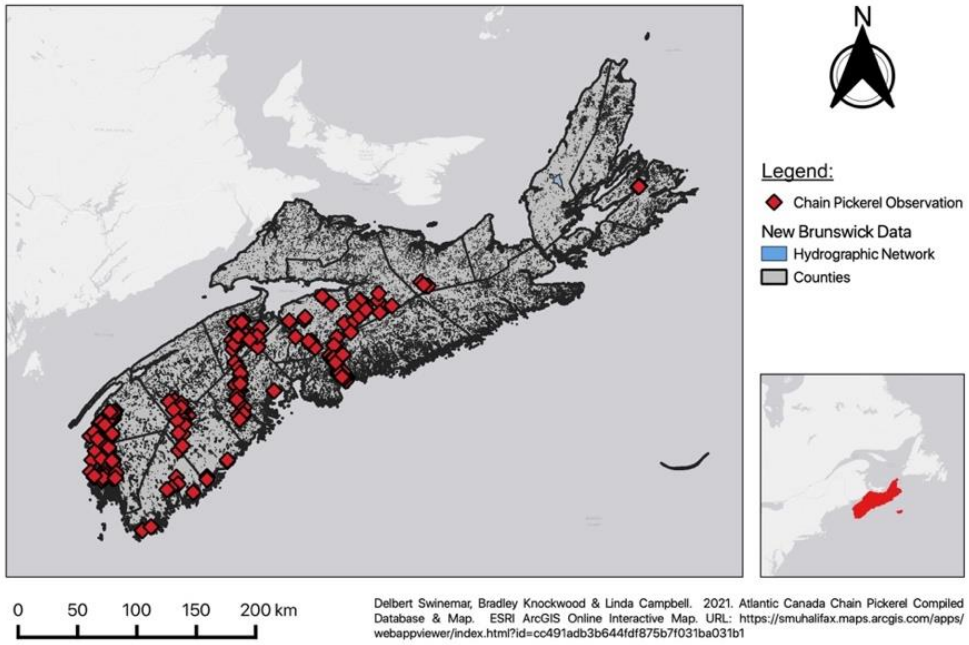

Figure 4: Distribution of E. niger in Nova Scotia. Total number of distinct water bodies with chain pickerel reports in 2021 = 201. (Swinemar et al., 2021).

New Brunswick Chain Pickerel Distribution

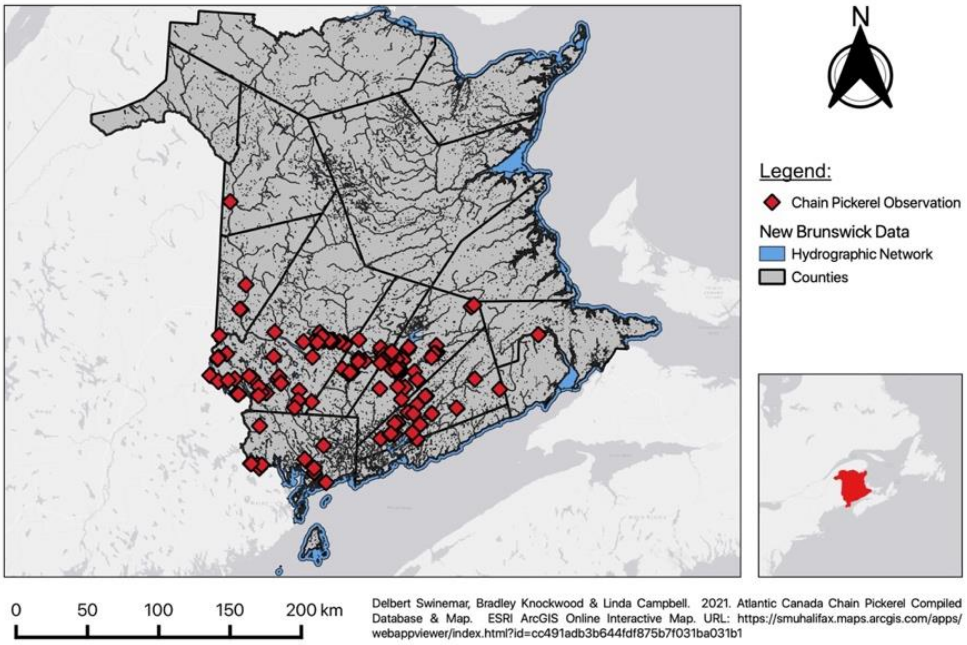

Figure 5: Distribution of E. niger in New Brunswick. Total number of distinct water bodies with chain pickerel reports in 2022 = 143. (Swinemar et al., 2021).

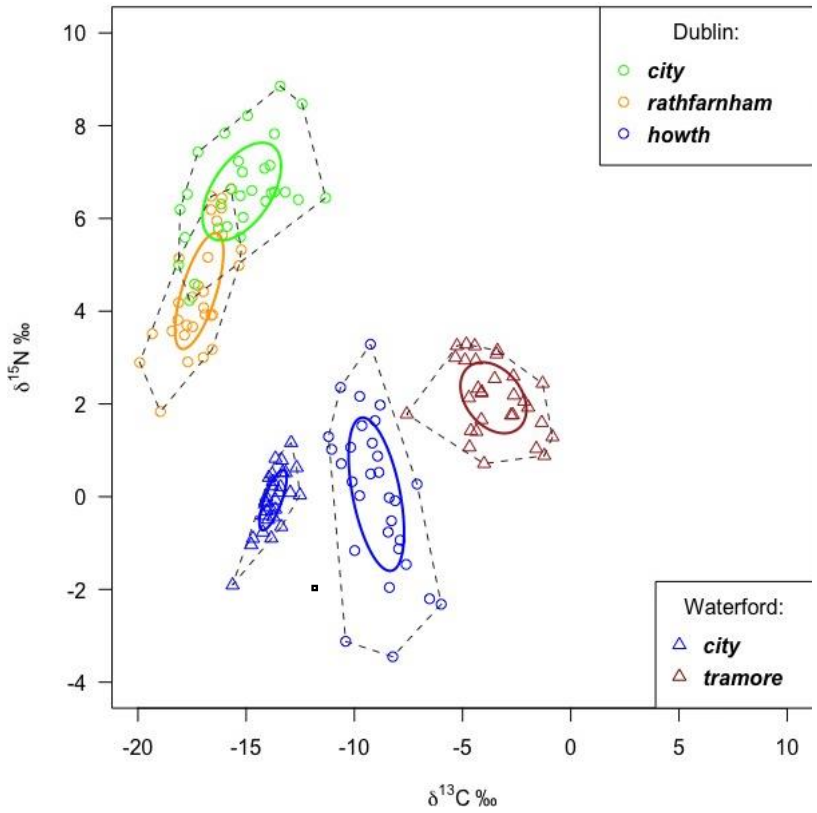

Figure 6: An example of a SIA bi-plot using randomly generated stable isotope data.

1.10 Tables

Table 1: Lakes with confirmed first reports of invasive E. niger *in KNPNHS. Data provided by Parks Canada (Parks Canada, 2019, 2020; D. Reid & D. Swinemar, personal communication, January 21, 2021).*

Location	Easting	Northing	Zone	Date Observed (yyyy-mm-dd)
Mersey River	324592	4910700	20T	2018-06-24
Kejimkujik Lake	322742	4918717	20T	2018-08-22
Peskowesk Brook	326223	4905285	20T	2018-08-23
Loon Lake	325380	4909960	20T	2018-08-20
Rogers Brook	322985	4919381	20T	2018-10-21
Peskowesk Lake	319772	4908636	20T	2018-10-23
Grafton Brook	324455	4916682	20T	2018-07-17
West River	316945	4917583	20T	2019-07-31
Grafton Lake	325932	4916962	20T	2019-10-22
Frozen Ocean Lake	313375	4924805	20T	2020-07-27
Mill Falls	323664	4923024	20T	2020-08-26
Big Dam West Lake	317933	4925814	20T	2020-10-14
Cobrielle Lake	321715	4909149	20T	2021-06-08

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Chapter 2: The shifting trophodynamics in four southern Nova Scotia lakes after the introduction of Chain Pickerel (*Esox niger*).

2.0 Introduction

Kejimkujik National Park and National Historic Site (KNPNHS) has a long and rich history of aquatic resource usage (Basquill et al., 2001; Clifford Drysdale, 1986). For centuries the Mi'kmaq peoples used the region extensively as a hunting and foraging ground, utilizing freshwater resources as part of their main food sources (Basquill et al., 2001; Morrison, 1977; Parker, 2016). After European settlers arrived and moved throughout eastern Canada, many sports fishermen visited the Kejimkujik region to take advantage of its abundant aquatic resources for decades (Morrison, 1977; Paine, 1967). The "sporting era" in Nova Scotia refers to a historical period when outdoor recreational activities like fishing were primarily pursued for leisure rather than subsistence or economic purposes (Morrison, 1977; Parker, 2012, 2016). Starting in the 1860-1870's the sporting era reached its peak by the 1880-1930's and ended by the 1950's (Morrison, 1977; Parker, 2012, 2016).

Chain Pickerel (*Esox niger*) were first reported for KNPNHS in June 2018 (Brake, 2020; Parks Canada, 2019, 2020). Already, only a few years after the introduction, reports are emerging that *E. niger* are causing declines in abundances of many native fish species within KNPNHS (Brake, 2020). Since the first report of chain pickerel in 2018, Parks Canada and a team of volunteer anglers have been closely monitoring the spread of this species throughout KNPNHS (*Table 1*). As a result, we now have a database of years of first reports for various lakes which is considered highly accurate. Therefore, we can leverage this database to design a

sampling framework to assess the changes of food webs corresponding with the length of time chain pickerel has been in the lake.

It is hypothesized that invasive *E. niger* remodel food web structure within invaded lakes by removing vulnerable prey types in a similar manner to other invasive *Esox* species (Brake, 2020; MacLeod, 2020; Mitchell et al., 2011). Chain Pickerel will undergo a distinct and consistent size-based dietary shift, with smaller fish less than 10 cm transitioning from benthic macroinvertebrates (BMI), especially Odonata nymphs (Hunter & Rankin, 1939; MacLeod, 2020; Meyers & Muncy, 1962; Mitchell et al., 2011; Raney, 1942) to larger fish species as they grow more than 10 cm (Gilhen & Pentz, 1974; Lee et al., 1980; Livingstone, 1950; MacLeod, 2020; Scott & Crossman, 1973).

This study utilized a four-lake framework based on the spread of *E. niger*. After the first introduction of *E. niger*, Parks Canada began to monitor and track the spread of *E. niger* through the connected KNPHS waterbodies By documenting its progression into new habitats, we were able to observe its invasion timeline and adapt a spatial approach to our research which includes different time lengths of invasion presence. These observations allowed us to design an experimental framework that captures key ecological changes associated with the species' establishment over time. Four lakes were selected based on their accessibility and the timeline of invasion: Loon Lake (first reported 2018, 4 years post-invasion), Grafton Lake (2019, 3 years post-invasion), Big Dam West Lake (2020, 2 years post-invasion), and Cobrielle Lake (2021 <1 year post-invasion). By utilizing this approach, we were able to approximate temporal change across our four study lakes.

Stable isotope analysis (SIA) with carbon ($\delta^{13}C$) and nitrogen ($\delta^{15}N$) isotope ratios can be used to characterize food webs and extrapolate the possible dietary preferences of *E. niger* (Fry,

2006; Post, 2002; Vander Zanden et al., 1999). Mixing models using Bayesian statistical analysis of $\delta^{13}C$ and $\delta^{15}N$ and are useful tools for assessing possible dietary contributions of multiple sources on a specific consumer (Phillips et al., 2014).

Here, we use a 4-lake experimental design with each lake having had chain pickerel for different times (0 years to 4 years of chain pickerel persence) to assess the possible temporal changes on food webs with stable isotope analyses. Our priority questions for this study are: (1) Are *E. niger* feeding preferably on any particular carbon source such as benthic invertebrates, native fish or pelagic zooplankton sources? (2) Is *E. niger* diet related to size class? (3) How are *E. niger* affecting food web structure over time?

2.1 Materials and Methods

Kejimkujik National Park and National Historic Site (KNPNHS), established in 1968 (Basquill et al., 2001) is located in southern Nova Scotia, with an area of 381 km^2 (44°21'50 "N, 65°18'08"W) (Figure 7). KNPNHS is characterized by a post-glacial landscape, marked by glacial erratics, eskers and drumlins, with numerous shallow stream, river and lake beds (Gimbarzevsky, 1975). All lakes in KNPNHS are highly dystrophic (brown water) and have moderate total organic carbon (TOC) because of the low buffering capacity of the soils and abundance of bogs and fens in the watersheds (Ginn et al., 2007). KNPNHS has the warmest mean annual temperatures in Eastern Canada (Watson 1974, Shaw 1997). This relatively moderate climate support unique disjunctive population of plant and animal species otherwise found further south in Massachusetts (Bleakney 1963, Drysdale 1986).

Four lakes with different times of first reports of *E. niger* were selected based on their accessibility and invasion timeline: Loon Lake (first reported 2018, 4 years post-invasion at time

of sampling), Grafton Lake (2019, 3 years post-invasion), Big Dam West Lake (2020, 2 years post-invasion) and Cobrielle Lake (2021, <1 year post-invasion) (Figure 8, Table 1, Table 2). All study lakes are considered oligotrophic or mesotrophic, polymictic, and highly acidic ($5.2 \leq pH \leq 6.2$) and range from 73.8 to 200 Ha (Table 2).

2.1.2 Native and Invasive Fish

A total of 266 fish samples were collected over a total of 213 hours sampling effort via the (KNPNHS) native fish monitoring program (Table 4) in Fall 2020 (September 24 to October 26, 2020) and Spring 2021 (June 8 to July 1, 2021). Two Alaska trap nets are set twice a year in Spring and Fall at same locations as a part of the native monitoring program (Figure 8) and left for 1 week with checks every 24 hours. Every individual fish caught was measured for total length, fork length, weight and other details were noted.

Selected fish were euthanized, stored in individual plastic food grade or sterile bags, placed in a cooler with lab-grade freezer gel packs, and immediately transposed back to the KNPNHS field lab. Prior to dissection, fish were weighed, total length and fork length were measured and a ~1cm^3 section of dorsal muscle was removed from every individual with a total length greater than 5 cm. Whole-body samples were used if the fish total length was less than 5 cm. The Canadian Council on Animal Care (CCAC) animal care protocols for our fish sampling was approved the Acadian University Animal Care Committee, protocol number 06-20. All sampling were conducted under a Parks Canada Agency permit number KNP-2020-36298.

2.1.2.1 Gut Content Data

Gut content data was collected for 82 *E. niger* individuals from all lakes. Individual *E. niger* stomach contents were removed, weighed and then the contents of 59 guts identified to highest possible taxonomic resolution. Some gut content was unidentifiable. Gut content proportions were determined by dividing the number of species observations respectively by the sum of all gut content for CP1 and CP2 (see below for cluster analyses).

2.1.3 Benthic Macroinvertebrates

Benthic invertebrates were collected from all 4 lakes during the same sampling period for fish trapping and was done near each Alaska net trapping site using rock flipping and a CABIN-style 400-micron mesh kick net. A total of 137 Benthic Macroinvertebrates (BMI) were gathered with a cumulative total of 231 hours sampling effort (Table 5) during the same Fall and Spring sampling sessions as the fish.

While the Order Odonata was the focus of BMI sampling efforts, with 102 Odonata were sampled, all macroinvertebrates were retained for identification and analyses. At the lab samples were removed and washed with distilled water to remove sediments. Several Odonata and other aquatic invertebrate identification guides were used to identify down to species level or highest possible taxonomy resolution (Clifford, 1991; May & Dunkal, 2007, 2007; Peckarsky, 1993; Tennessen, 2021), while other macroinvertebrates were typically identified to Order or Genus level.

2.1.4 Zooplankton

A total of 16 bulk zooplankton samples were gathered from all 4 lakes. Pelagic zooplankton samples were collected from a boat at the deepest site in each study lake. A 30.48-cm tow net was hauled to the surface at a rate of ~0.5 m/s, (Nordin et al., 1997) and repeated 13 times at each zooplankton sampling site.

In the lab, zooplankton samples were size fractioned by filtering through 243-µm and 53-µm Nitex mesh and fractionated by mesh size (Table 6). One sample per size fraction from each study lake was preserved in 10% neutral buffered formalin and sent to IdentaZoop in Ontario Canada. A total of 1,500 zooplankton were identified making up 23 species across the four study lakes (Appendix A: Raw Data), with the most common species being *Eubosmina longispina* and the least common species being *Leptodiaptomus siciloides*, *Daphnia ambigua*, *Chydorus sphaericus*, and *Alona spp*. for all size classes (Table 6). The three most common species in the 243-µm size class were *Holopedium gibberum*, *Eubosmina longispina* and *Daphnia catawba* (Table 6). For the 53-µm Calanoid copepods, *Epischura copepodid* and *Eubosmina longispina* were the most common taxa identified (Table 6).

2.1.5 Sample Processing

All samples were placed in 1 N hydrochloric acid- cleaned 20-mL glass scintillation vials, and dried for a period of 24-48 hours at 60°C. After drying was completed, samples were weighted again to determine dry weight. Once all samples were dried, all samples were sent to the Stable Isotopes in Nature Laboratory (SINLab) to be ground and analyzed via the Continuous Flow-

Isotope Ratio Mass Spectrometry (CF-IRMS) (University of New Brunswick, Fredericton, New Brunswick) following their analytical methodologies for $\delta^{13}C$ and $\delta^{15}N$ analyses (Stable Isotopes in Nature Laboratory, 2021).

2.2 Data Analysis

The raw data included $\delta^{15}N$ and $\delta^{13}C$ values, %C, %N and C:N ratios for each sample. Fish were labeled by 2-letter capital letter codes based on their common names, BMI were labeled lower-case codes based on the lowest identified taxonomic identification, and all zooplankton were labeled as ZO (Table 7).

Data analysis was run using R version 4.4.0. MixSIAR version 3.1.12 was downloaded directly the GitHub repository (Stock & Semmens, 2016). NbClust version 3.0.1 was downloaded from CRAN (Charrad et al., 2014). tRophicPositon version 0.8.0 was downloaded directly the GitHub repository (Quezada-Romegialli et al., 2018).

2.2.1 Lipid Correction

C:N ratios in fish and invertebrates were tested for differences among species within each study lake. If C:N varied significantly, then the corresponding δ13C data were numerically adjusted (e.g. "corrected") for lipid content using the Kiljunen model as outlined in Chapter 1 (Kiljunen et al., 2006). The isotopic difference between protein and lipids (D) was defined as D = 7.018 ± 0.263 (Kiljunen et al., 2006). The intersection on the x-axis (I) was defined as I = 0.048 (Kiljunen et al., 2006). Lipid corrected values were stored separately alongside $\delta^{13}C$ as $\delta^{13}C_{lipid.}$

2.2.3 Cluster Analysis

Cluster analysis was performed on raw unadjusted $\delta^{15}N$ and $\delta^{13}C$ *E. niger* values across all study lakes. The NbClust package was used to partition *E. niger* stable isotope data into an optimal number of distinct clusters k, see Chapter 1.5.3 Cluster Analysis. *E. niger* $\delta^{15}N$ and $\delta^{13}C$ data were assigned clusters using the NbClust k-means cluster assignment algorithm. A new data column was created in the stable isotope dataset indicating which cluster each *E. niger* belonged to. The centroid points $\Psi(\delta^{13}C, \delta^{15}N)$ and Ω(TL) and SD uncertainties were determined using material covered in Chapter 1.5.3 Cluster Analysis. Midpoints of every cluster in δ-space were determined by averaging $\delta^{13}C$ and $\delta^{15}N$ values for every individual per cluster. Average total lengths for every cluster were determined by averaging the total lengths for every *E. niger* assigned to each cluster respectively.

2.2.4 Mixing Model (MixSIAR)

Prior to running MixSIAR, raw data with *E. niger* cluster assignment was regrouped in the following way: all native fish were grouped into “fish”, Odonata were grouped into “odo” and zooplankton were renamed “ZO” for each study lake, Table 10. These groupings were considered sources in the 3-source model. *E. niger* were grouped by lake and then by cluster assignment depending on the outcome of cluster analysis and the optimal number of clusters k. Consumer means were calculated by averaging $\delta^{15}N$ and $\delta^{13}C$ values for each split by lake. An uninformative alpha prior was used for the model assuming that every source initially has an equal chance of being consumed by *E. niger* ($\alpha = (1, 1, 1)$).

Trophic $\delta^{15}N$ enrichment factors (TEF) were used for fish, odo and ZO respectively. $\delta^{15}N$ TEF of 1.4 ± 0.5 was used for odo and ZO, while 3.4 ± 0.5 was used for fish (McCutchan et al., 2003; Vander Zanden et al., 1999). $\delta^{13}C$ TEF of 0.3 ± 0.5 was used for odo and ZO while 1.3 ± 0.5 was used for fish (McCutchan et al., 2003; Vander Zanden et al., 1999).

There were challenges handling the data for certain lakes which required some decisions. Only one bulk zooplankton sample could be gathered for Grafton Lake, and to run MixSIAR at least two samples are required. Given the high consistency of pelagic zooplankton SIA values among lakes, we used the mean of the standard deviation for all other zooplankton samples from other study lakes were used as the $\delta^{13}C$ and $\delta^{15}N$ means for the Grafton Lake zooplankton sample. Only one *E. niger* was caught and sampled for $\delta^{13}C$ and $\delta^{15}N$ at Cobrielle Lake and only 1 large CP1 captured in Big Dam West Lake (Table 9). As a result, CP1 and CP2 for Cobrielle Lake and CP1 for Big Dam West Lake were not used in the MixSIAR models for those 2 lakes due to the limited consumer sample size.

The MixSIAR model outputs a table of probable source contributions to each consumer group, corresponding standard deviation for each study lake. Model quantitative results are reported as likely proportions with SD as uncertainties, ranging from 0 (no contribution) to 1 (100% contribution). MixSIAR outputs posterior density plots depicting likely proportions of each source's contribution to each consumer group respectively. The x-axis represents the proportion of each source's contribution to the consumer's isotopic signature, ranging from 0 (no contribution) to 1 (100% contribution). The y-axis shows the probability density for different proportions of the source contributions. Peaks indicate the most likely contributions for each source to each consumer.

2.2.5 Trophic Position Modelling

Trophic positions of distinct species groups were determined using the tRophicPosition package (Quezada-Romegialli et al., 2018). Bayesian modeling methodologies are utilized for the precise calculation of consumer trophic positions utilizing $\delta^{15}N$ and $\delta^{13}C$ stable isotope data, accommodating scenarios with either one or two baselines.

tRophicPosition allows for adjustment of varying baselines. Due to the very low number of freshwater mussels in the acidic KNPNHS lakes, zooplankton and Odonata were selected as the primary baseline organisms. Trophic positions (TP) for every species group were calculated using the R library rTrophicPosition (Table 12 and Figure 14).

2.3 Results

2.3.1 Raw Data Observation

The stable isotope biplots indicate that *E. niger* are feeding from two different consumer classes in the two lakes with longest presence of chain pickerel (Loon and Grafton Lakes) (*Figure 10*). In those two lakes, *E. niger* with high $\delta^{15}N$ values ($\delta^{15}N \sim 9‰$) appear near native fish species. *E. niger* with low $\delta^{15}N$ values ($\delta^{15}N \sim 5‰$) appear near BMI species, specifically Odonata. Additionally, *E. niger* $\delta^{15}N$ values were plotted with total length (TL) for Loon, Grafton and Dam West Lakes. The $\delta^{15}N$ vs TL plots suggest there are two distinct clusters of *E. niger* in those post-invasion study lakes (Table 8 and *Figure 10*).

Layman food web metrics for Big Dam West Lake (2020) indicated the widest food web with a carbon range of 6.6‰ while Loon Lake (2018) had the smallest (3.4‰) (Table 8 and

Figure 9). Loon Lake had the longest food web with a nitrogen range of 5.9‰ (Table 8 and Figure 9). The distance to centroid (CD) was similar for all study lakes, the trophic diversity within food web is relatively uniform (Table 8 and Figure 10). Food web total area (TA) was greatest in Big Dam West Lake (Table 8 and Figure 10). Big Dam West Lake had the highest overall nearest neighbor distance (Table 8 and Figure 10).

2.3.2 Cluster Analysis

NbClust was used to determine the optimal number of *E. niger* clusters (k). The majority of the 30 NbClust clustering indices voted for k = 2 as an optimal number of *E. niger* clusters. *E. niger* were grouped into two distinct clusters in δ-space (Figure 10 and Figure 11). *E. niger* assigned to cluster 1 and cluster 2 coded as CP1 and CP2 respectively (Figure 12). The centroid of CP1 and CP2 with SD were found to be -30.2 ± 1.2‰, 5.4 ± 0.6‰ and -29.2 ± 1.0‰, 8.8 ± 0.7‰ respectively. CP1 had a $\delta^{15}N$ and $\delta^{13}C$ range of 4.1‰ ≤ $\delta^{15}N$ ≤ 6.4‰ and -30.1‰ ≤ $\delta^{13}C$ ≤ -28.9‰ respectively. CP2 had a $\delta^{15}N$ and $\delta^{13}C$ range of 7.1‰ ≤ $\delta^{15}N$ ≤ 10.4‰ and -30.6‰ ≤ $\delta^{13}C$ ≤ -25.7‰ respectively. Average TL of *E. niger* belonging to CP1 and CP2 was 5.69 ± 1.50cm and 37.13 ± 11.65cm respectively.

The centroid point between clusters CP1 and CP2 in δ-space, denoted as Ψ is where *E. niger* has an equal chance of being assigned to either CP1 or CP2 (Kassambara, 2017). Therefore Ψ represents a transition point between the two clusters and can be interpreted as the point in δ−space that *E. niger* undergoes a dietary shift, transitioning form CP1 to CP2. Ψ was found to be -29.7 ± 1.8‰, 7.1 ± 0.8‰ in δ−space (Figure 12).

The TL centroid Ω between clusters CP1 and CP2 represents a transition point between two TL clusters and was found to be 21.41 ± 2.24cm. The average TL of CP1 and CP2 respectively were 5.69 ± 1.50cm and 36.42 ± 12.63cm with SD. The total length ranges of *E. niger* assigned to CP1 and CP2 were 4.2cm ≤ *TL* ≤ 10.9cm and 5.5cm ≤ *TL* ≤ 58.6cm respectively.

2.3.3 MixSIAR Model

MixSIAR was used to determine the dietary contributions of three sources for two *E. niger* consumer groups, CP1 and CP2. Consumer means for CP1 and CP2 are shown in (Table 9). The MixSIAR model output for CP1 suggests a dietary contribution corresponding with Odonata make up a 0.53 ± 0.07 and 0.85 ± 0.09 proportion of *E. niger* diet for Loon Lake (2018) and Grafton Lake (2019) respectively (Table 11 and Figure 13). Unfortunately, there were not enough CP1 individuals caught in Big Dam West (2020) and Cobrielle (2021) Lakes to perform the analysis for these cases.

MixSIAR modeling for CP2 suggest that dietary contribution corresponding with small fish consists of 0.71 ± 0.04, 0.74 ± 0.02 and 0.77 ± 0.05 of *E. niger* diets for Loon, Grafton and Big Dam West Lake respectively (Table 11 and Figure 13). There were not enough CP2 individuals caught in Cobrielle Lake to perform the analysis for this case.

2.3.4 Gut Content

Gut content data was collected for 82*E. niger* and 59 guts had sufficient content for taxonomic analyses. Odonata made up the highest proportion of CP1 diet across all study lakes

with an average proportion of 0.55 (Table 13). Fish made up the highest proportion of CP2 diet across all study lakes with an average proportion of 0.85 (Table 14). Yellow and white perch made up the highest proportion of CP2 diet (Table 14).

2.3.5 Trophic Position

Using zooplankton- and BMI-adjusted baselines, the trophic position (TP) of every species grouping was calculated (Table 12 and Figure 14). The mean TP of all native fish species decreased on average by 0.43 ± 0.06 when comparing more recently-invaded lakes, Cobrielle (2021) and Big Dam West (2020) Lakes, with those lakes with chain pickerel for longer times, Grafton (2019) and Loon Lakes (2018) (Table 12). The mean TP of CP1 were found to decrease by 0.58 ± 0.27 when comparing Cobrielle and Big Dam West Lakes with Grafton and Loon Lakes (Table 12). The mean TP of CP2 were found to decrease by 0.21 ± 0.34 when comparing Cobrielle and Big Dam West Lakes with Grafton and Loon Lakes (Table 12). The mean TP of White Perch and Yellow Perch decreased the most (0.75 ± 0.24 and 0.64 ± 0.30 respectively) when comparing Cobrielle and Big Dam West Lakes with Grafton and Loon Lakes (Table 12 and Figure 14). The mean TP of all Odonata species decreased on average by 0.41 ± 0.44 when comparing Cobrielle and Big Dam West Lakes with Grafton and Loon Lakes (Table 12).

2.4 Discussion

Our work focusing on 4 lakes in KNPNHS indicate that how long *E. niger* is present in lakes is strongly associated with food web changes, with the lakes having had *E. niger* for 3-4 years having the most significant changes compared to those which had chain picker *E. niger* for

only 1 to 2 years. This is in line with studies in Nova Scotia and elsewhere have indicated that the presence of *E. niger* in lakes is consistently associated with decreased native fish and benthic macroinvertebrate communities (Alexander et al., 1986; Gilhen & Pentz, 1974; Livingstone, 1950; Mitchell et al., 2011). A review of *Esox* spp and *E. niger* studies elsewhere indicate that invasive *E. niger* remodel the food web structure of lakes by removing vulnerable prey types resulting in a loss cyprinid fish species, truncation of fish body size distribution and likely a change in lake function (Mitchell et al., 2011). Several studies have shown that *E. niger* prey heavily on Odonata and native fish species resulting in an overall decrease in abundances (Brake, 2020; MacLeod, 2020; Mitchell et al., 2011).

We used the MixSIAR program to estimate the dietary contributions of fish, Odonata, and zooplankton to *E. niger* in the smaller CP1 and larger CP2 groups in all 4 lakes. In lakes where *E. niger* has been present for 3 to 4 years, there are two distinct feeding groups based on size class indicating a dietary transition at (-29.7 ± 1.8‰, 7.1 ± 0.9‰) in δ-space happening when the fish are around 21.41 ± 2.24 cm total length. A similar dietary transition has been shown to occur in other studies investigating the diet of *E. niger*.

It has been a matter of common knowledge that *E. niger* and other species of *Esox* follow a bifurcated size class distribution. Hunter and Rankin observed this behaviour in specimens of *E. niger* (Hunter & Rankin, 1939). *E, niger* were divided into two feeding groups based on their diet, those with TL < 6in (TL <15cm) and those with TL > 6in (TL > 15cm) (Hunter & Rankin, 1939). Those with TL < 6in were found to mostly feed on aquatic invertebrates, while those with TL > 6in were found to predate mostly on smaller fish species (Hunter & Rankin, 1939). Scott and Crossman observed a similar dietary bifurcated distribution in diet for *E. niger* (Scott & Crossman, 1973).They observed a distinct separation into two distinct niches, those that feed

primarily on aquatic invertebrates when TL < 10.2 – 15.2cm, and those that feed mostly on smaller native fish species when TL > 10.2 – 15.2cm (Scott & Crossman, 1973). More recent MacLeod was able to show that *E. niger* have two distinct feeding groups based on gravimetric analysis; an invertebrate-dominant group and fish-dominant group (MacLeod, 2020). In the invertebrate-dominant group invertebrates occurred in 88% of *E. niger* stomach samples, contributing 13.9% of their diet by weight (MacLeod, 2020). Dragonfly nymphs (Odonata) were the most frequently consumed invertebrates, comprising 89.7% of the invertebrate category (MacLeod, 2020). In the fish-dominant group fish constituted 76% of the total stomach content weight and were found in 30% of the samples (MacLeod, 2020). Among the fish prey, Atlantic Salmon (*Salmo salar*) smolts made up 46%, followed by White Perch (*Morone americana*) at 31.4% (MacLeod, 2020). The average TL of CP1 and CP2 respectively were 5.69 ± 1.50cm and 36.42 ± 12.63cm with SD. The total length ranges of *E. niger* assigned to CP1 and CP2 were 4.2cm ≤ TL ≤ 10.9cm and 5.5cm ≤ TL ≤ 58.6cm respectively. The results of my study are consistent with findings from other researchers in similar studies.

Our mixing model results indicate that that the smaller CP1 groups feed mostly on Odonata with dietary contributions of 0.65 ± 0.07% and 0.95 ± 0.09% for Loon and Grafton Lakes respectively (average of 0.8 ± 0.11%). Results for the larger CP2 indicate a larger proportion of native fish with dietary contributions of 0.71 ± 0.04%, 0.74 ± 0.02% and 0.77 ± 0.05% for Loon, Grafton, and Big Dam West Lakes respectively (average of 0.74 ± 0.07%).

Gut content data provide additional support for MixSIAR inferred diets of CP1 and CP2 *E. niger*. Gut content data for CP1 *E. niger* suggests that CP1 feed mostly on Odonata with a proportion of 0.55 (Table 13). Gut content data for CP2 *E. niger* suggests that CP2 feed mostly

on native fish, specifically, yellow perch and white perch with proportions of 0.32 and 0.25 respectively (Table 14). These results demonstrate the trophic adaptability of *E. niger*.

It is well established that smaller (younger) *E. niger* feed primarily, while larger (older) pickerel are primarily piscivorous (Foote & Blake, 1945; Hunter & Rankin, 1939; Raney, 1942). Mitchel et al. found that smaller *E. niger* diet was dominated by invertebrates, especially insects such as dragonfly and mayfly nymphs, while larger *E. niger* diet shifted to being primarily piscivorous (fish-eating) (Mitchell et al., 2011). MacLeod found that invertebrates make up 88% of smaller *E. niger* diet while native fish make up 76% of larger *E. niger* diet (MacLeod, 2020). Hunter and Ranken found that the smaller size class of *E. niger* diet consisted of 67.4% invertebrates while the larger size calas diet consisted of 52.4% smaller fish (Hunter & Rankin, 1939). The MixSIAR model and gut content results of my study are consistent with findings from other researchers in similar studies.

Esox follow a linear and predictable growth curve (Lorantas, 2018; Lorenzoni et al., 2002; Scott & Crossman, 1973). Several papers reference a period of rapid growth in juvenile *E. niger*, reaching ~10cm by the end of the first summer, and growing at a rate of ~6cm/year during year two and moving forward (Foote & Blake, 1945; Raney, 1942; Scott & Crossman, 1973; Underhill, 1949). The sudden jump in total length seen in Figure 11 may be a result of rapid growth as described by Foote & Blake (1945), Raney (1942), Scott & Crossman (1973), and Underhill (1949), or due to study design and the time of sampling. *E. niger* were sampled in Fall 2020 (September 24 to October 26, 2020) and Spring 2021 (June 8 to July 1, 2021), and several factors could account for the gap between these two sampling periods, including:

Seasonal Variation in Growth:

Fish growth often varies seasonally, with faster growth rates observed during warmer Summer months when metabolic activity and food availability are at their peak (Scott & Crossman, 1973). For *E. niger*, similar patterns of growth during warmer months have been reported (Scott & Crossman, 1973).

Sampling Timing:

The timing of sampling during the Fall and Spring may have coincided with different stages in the fish's life cycle or seasonal growth patterns (Eerola et al., 2024; Scott & Crossman, 1973). Fish often grow more rapidly during the warmer months, so sampling before and after these periods might capture different size distributions (Eerola et al., 2024; Scott & Crossman, 1973).

Sampling Bias:

Different sampling methods have biased captures based on fish size or behavior (Lake et al., 2001; Nordin et al., 1997). In general, smaller fish tend to avoid capture in certain gear, whereas larger fish may be more likely to be caught in other gear (Lake et al., 2001; Nordin et al., 1997). This could result in a disproportionate representation of certain sizes influencing perceived growth rates.

Fish Age and Development:

As fish age their growth rate often decreases, the observed jump in size could coincide with a particular age or growth stage. Studies on the growth patterns of *E. niger* indicate that rapid juvenile growth that slows once fish reach a certain size (Scott & Crossman, 1973; Raney, 1942).

The growth rate in juvenile fish is often much higher in the early years compared to later stages (Scott & Crossman, 1973; Raney, 1942).

Some of these factors may contribute to the observed growth patterns seen in Figure 11. Further research or more frequent sampling following the same methodologies used in this study could help clarify the underlying causes.

2.4.1 *E. niger* and Odonata

The results of the mixing model and gut content data suggest that Odonata constitute the highest proportion of CP1 diet. The order Odonata consists of dragonflies (Anisoptera) and damselflies (Zygoptera) (Tennessen, 2021). Odonata appear in nearly every type of water body, from seeps and streams to ponds, lakes, and even some temporary pools are inhabited by some species of Odonata (Tennessen, 2021). Odonata nymphs are primarily lentic feeders spending most of their time hunting for food amongst rocks and aquatic vegetation in the littoral zone of lakes (Paulson, 2011; Peckarsky, 1993; Tennessen, 2021).

Odonata nymphs consistently are main food of certain *Esox* species. It is known that *E. lucius* and *E. americanus* prey on weed-dwelling Odonata more often than burrowers (Tillyard, 1917). Odonata nymphs are mostly weed-dwellers typically being found clinging onto submerged vegetation, on rocks and submerged logs (Corbet, 1999; Paulson, 2011; Tennessen, 2021). They will often crawl up on anything that may be convenient when metamorphosizing, often emergent vegetation (Tillyard, 1917). Odonata nymphs are susceptible to predation by predatory fish species such as trout and *Esox sp.* as the nymphs climbing along the stems of macrophytes are often exposed (Kidd et al., 2011; Tillyard, 1917). Odonata habitat overlap with

E. niger likely plays a role in this preference, as *E. niger* are well documented opportunistic predators, they may easily take advantage of Odonata vulnerability while feeding or preparing for metamorphosis (Scott & Crossman, 1973; Tillyard, 1917).

2.4.2 *E. niger* and Native Fish

Mixing model results and gut content data suggest that cluster CP2 contain *E. niger* that relied mostly on native fish food sources. Several studies have reported *E. niger* undergo a distinct dietary shift transitioning from a reliance on BMI (CP1) to smaller fish species (CP2) hypothesized . The approximate total length for the transition varies depending on the study. Many sources report a dietary transition occurring at TL of 10 – 15cm when *E. niger* are 1 year old (Coffie, 1998; Foote & Blake, 1945; Hunter & Rankin, 1939; Meyers & Muncy, 1962; Scott & Crossman, 1973). *E. niger* dietary transition from BMI to fish depends on source availability (Coffie, 1998; Scott & Crossman, 1973). Due to their generalist behaviour, if fish are scarce, *E. niger* will continue to rely on BMI well past 15cm TL (Coffie, 1998; Scott & Crossman, 1973).

Cluster analysis results suggest that *E. niger* undergo a dietary transition at a greater TL than cited in literature. It was shown that *E. niger* likely transition from CP1 into CP2 at a total length of 19 – 24cm. Mixing models results suggest that *E. niger* with TL less than 19cm are likely feeding on Odonata. While mixing models results suggest *E. niger* with TL greater than 24cm are likely feeding on native fish.

E. niger growth rate vary considerably from place to place (Scott & Crossman, 1973). In more productive systems *E. niger* grow and mature more quickly than those in less productive (Coffie, 1998; Scott & Crossman, 1973). All study lakes are considered oligotrophic or mesotrophic, polymictic, and acidic (pH < 6). It is likely that a greater transition total length is

directly associated with the overall productivity of study lakes (Wyn et al., 2009, 2010). The transition TL is likely specific to the KNPNHS population of *E. niger* and not the overall Nova Scotia population. Lakes within KNPNHS have relatively similar productivity and water quality aspects, this statement is not true for every invaded lake and river throughout Nova Scotia (Clifford Drysdale, 1986; Gimbarzevsky, 1975; Joseph Kerekes & Peter Schwinghamer, 1973; Wyn et al., 2009, 2010).

2.4.3 Management Recommendations

E. niger in KNPHS lakes are here to stay, there are no practical means of completely eradicating this species from the large and complex interconnected lake and river systems in the park. Chemical fish-specific toxins such as rotenone are available (Dalu et al., 2015), but fish-specific chemical toxins are often only successful in small closed systems (Dalu et al., 2015), and therefore are not recommended for the highly interconnected waterbodies in the Park. Establishing early detection and rapid response (EDRR) protocols is imperative for minimizing or prevent invasive species impact within KNPNHS and other protected areas (Reaser et al., 2020). It is recommended that Parks Canada establish EDRR watershed-level invasive species monitoring and/or a freshwater monitoring protocols in a buffer zone around the KNPNHS boundaries, with monitoring taking places in connected lakes and rivers both outside and inside the KNPNHS b in close collaboration with local non-government, provincial government, and other community partners. It was known for decades that *E. niger* were in adjoining waterbodies, and if an early-detection system were in place, *E. niger* may have been detected earlier prior to invading lakes inside the KNPNHS.

Monitoring sites should be established in collaboration with partners at sites along major waterways flowing into and out of the KNPNHS. Multiple monitoring sites be established between 10-50km upstream and downstream of major watercourses; a monitoring site at 10km, another at 20km and so forth (Britton et al., 2011; Gallardo et al., 2016). At selected monitoring sites, a combination of environmental DNA (eDNA), visual surveys, electroshocking and fish nets should be used to detect the presence of aquatic invasives (Fonseca et al., 2023; Guo et al., 2024). Once detected, local stakeholders and the public should be notified immediately. Additional boat inspections should be enforced within KNPNHS by park wardens and in surrounding areas by provincial game wardens to prevent further spread. Boat ramps and other water access points should be temporarily closed.

Physical controls such as barriers may be set up to prevent detected invasives from spreading to connected water bodies (Krieg & Zenker, 2020; Mozzaquattro et al., 2020). Non-physical barrier such as CO_2 barriers have been gaining attention as effective measure for deterring invading aquatic species (Suski, 2020). Electric barriers have also gained attention and popularity (Layhee et al., 2016). Targeted removal efforts using electrofishing should be conducted by local fish and wildlife departments in coordination with Parks Canada to reduce the population. Local fishing regulations should be adjusted to encourage harvesting invasive species and to limit transport of live invasives between water bodies. Boat decontamination stations should set up within and outside KNPNHS at common boat launching sites to prevent spread by recreationists. Anglers should be encouraged to report any sightings. Research on new invasive species should be conducted to determine optimal removal/mitigation strategies.

Implementing stricter Clean Drain Dry protocols for boats entering the KNPHS areas as well as boats using the waterbodies within the buffer zone around the Park should be considered

(Department of Fisheries and Oceans Canada, 2021). A regular ongoing public awareness campaign as well as designated sites for cleaning watercraft would encourage all park visitors to clean, drain, and dry their equipment (boats, fishing gear, kayaks) before using the waterbodies. There should be a questionnaire and/or a written acknowledgment that visors are to follow clean, drain, and dry protocols prior to entering the park.

It is not too late to prevent other invasive species from moving into the Park and the buffer zone around the Park. For example, Marbled crayfish (*Procambarus virginalis*) have recently been confirmed in Yarmouth County and pose a threat to surrounding watershed (Maciaszek et al., 2022; Vogt et al., 2015). *P. virginalis* reproduce through parthenogenesis, and therefore a single introduced individual can rapidly multiply and take over an entire freshwater system (Scholtz et al., 2003). *P. virginalis* may have detrimental effects on the native invertebrates, amphibians, and fish, and alter the state of the invaded freshwater system (Maciaszek et al., 2022; Vogt et al., 2015). The Chinese mystery snail (*Cipangopaludina chinensis*) is present in Halifax County and other parts of the province (Kingsbury, 2021). *C. chinensis* affects the diversity of algae growing in its habitat, alters the water quality, and reduces the food supply for native snail species (Kingsbury, 2021). Zebra mussels (*Dreissena polymorpha*) is now found in the St. John's River in New Brunswick (New Brunswick Invasive Species Council, 2022) and presents a significant risk to Nova Scotia waterbodies. Therefore, chain pickerel presents a cautionary note, and with the potential arrival of new non-indigenous species, is worth investing in watershed level monitoring and implementation of strict Clean Drain Dry protocols within Park boundaries and in a buffer zone around the Park through partnerships in order to preserve ecological integrity, cultural heritage and traditional knowledge.

2.5 Conclusion

E. niger were found to occupy two distinct clusters in δ-space, CP1 (4.2cm ≤ TL ≤ 10.9 cm) and CP2 (20.2 cm ≤ TL ≤ 58.6 cm). Mixing model results and gut content data suggest CP1 *E. niger* diet is mostly constituted by Odonata while CP2 is mostly constituted by native fish, with average proportion of 0.65 ± 0.06 and 0.74 ± 0.02. Gut content proportions further suggest that CP2 *E. niger* feed primarily on yellow and white perch with overall gut content proportions of 0.32 and 0.25. Overall, this exemplifies the trophic adaptability of *E. niger* within study lakes.

Layman food web metric modelling suggest that the overall food web structure remains unchanged even in high-invasion scenarios. However, each fish species' trophic positions do change with the invasion length of time. Native top-trophic predatory fish species yellow perch and white perch appear to have decreased most significantly in TP when comparing Cobrielle and Big Dam West Lakes with Grafton and Loon Lakes. All native fish species sampled in this study were shown to have decreased TP post-invasion. Odonata sampled were also shown to have decreased TP post-invasion.

This study was designed to answer several questions aimed at understanding the impact of invasive *E. niger* within KNPNHS. Mixing model results and gut content data suggest smaller *E. niger* (CP1, see below) are feeding primarily on Odonata, while larger *E. niger* (CP2) are feeding primarily on native fish sources. There is a definite size class difference in feeding habits, as all *E. niger* in the 4 lakes were assigned into two non-overlapping clusters in delta space. Cluster CP1 contain *E. niger* with 4.2cm ≤ TL ≤ 10.9 cm, CP2 20.2 cm ≤ TL ≤ 58.6 cm. Furthermore, it was found that *E. niger* transitioned from feeding mostly on Odonata to native fish at a total length of 21.41 ± 2.24cm. *E. niger* is not significantly affecting the whole food web structure within 4 years because with Layman metrics indicating that overall food web structure

does not change significantly within a 3-4 year post invasion timeframe, however, the native fish and Odonata species trophic positions were found to decrease significantly post-invasion.

2.6 Figures

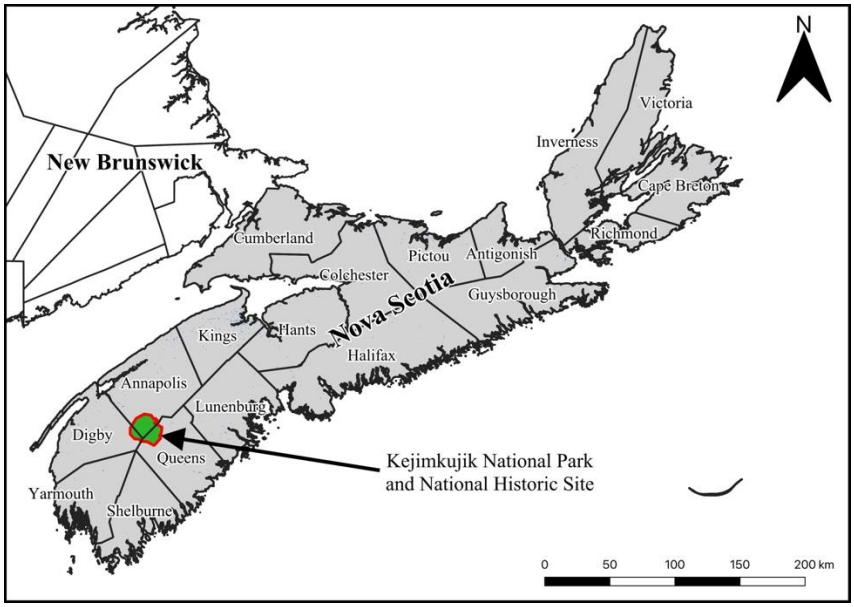

Figure 7: Kejimkujik National Park's location in Nova Scotia, Canada. Nova Scotia's County lines are indicated.

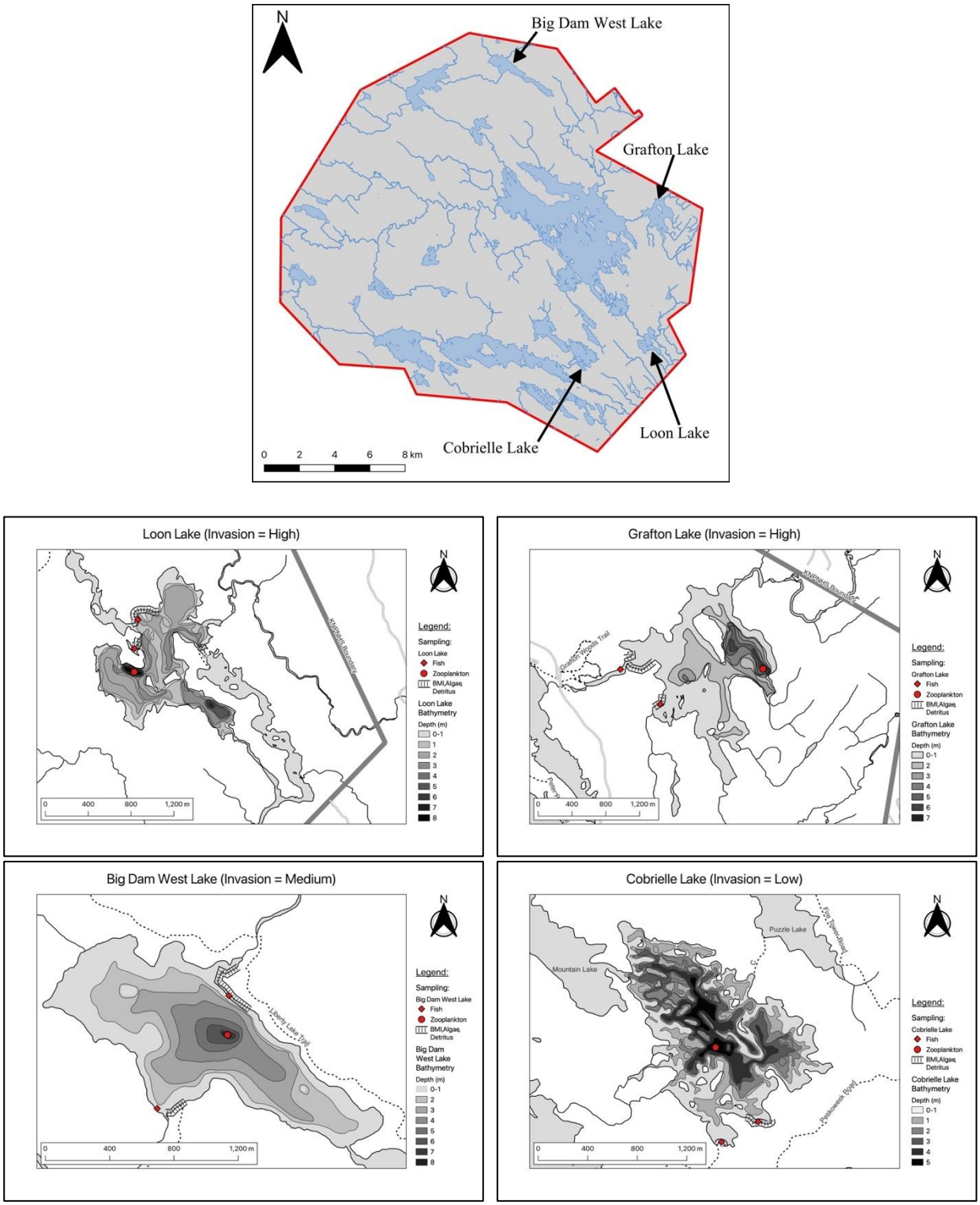

Figure 8: Top map provides an overview of Kejimkujik National Park and National Historic Site, the four study lakes (Loon Lake, Grafton Lake, Big Dam West Lake and Cobrielle Lake) are indicated and labeled. Maps top left to bottom right depict each study lake along with respective sampling sites; zooplankton, native fish, benthic macroinvertebrate, also depicted is bathymetric lake data (Joseph Kerekes & Peter Schwinghamer, 1973).

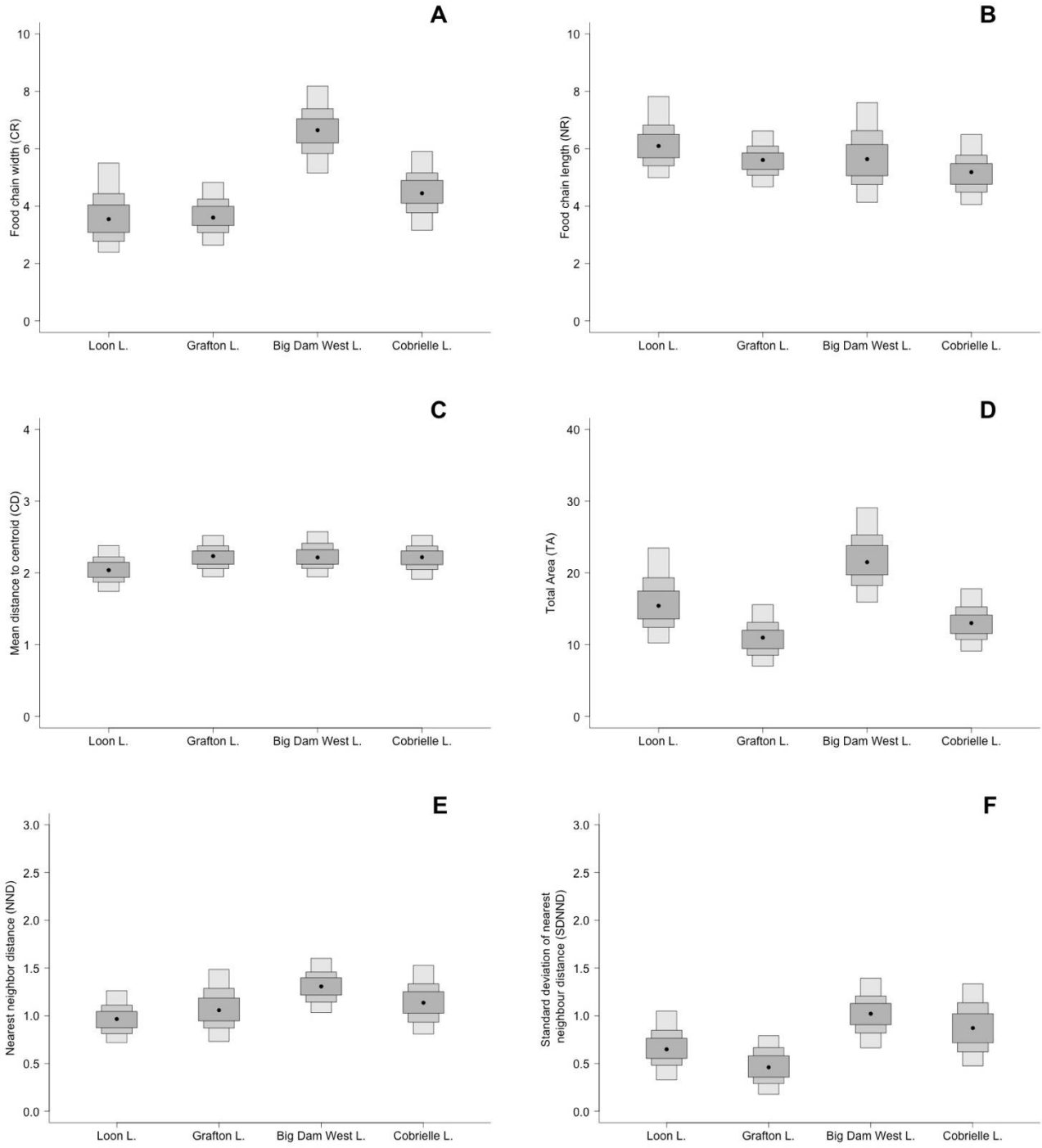

Figure 9: Density plots depicted in fig. 3A, 3B, 3C, 3D, 3E, 3F for the credible intervals of the Carbon Range (CR), Nitrogen Range (NR), Mean Distance to Centroid (CD), Total Area (TA), Nearest Neighbour Distance (NND) and Standard Deviation of Nearest Neighbour Distance (SDNND) of consumer data grouped by family for each sampling. Black dots represent the mode, and boxes present the 50%, 75% and 95% credible intervals.

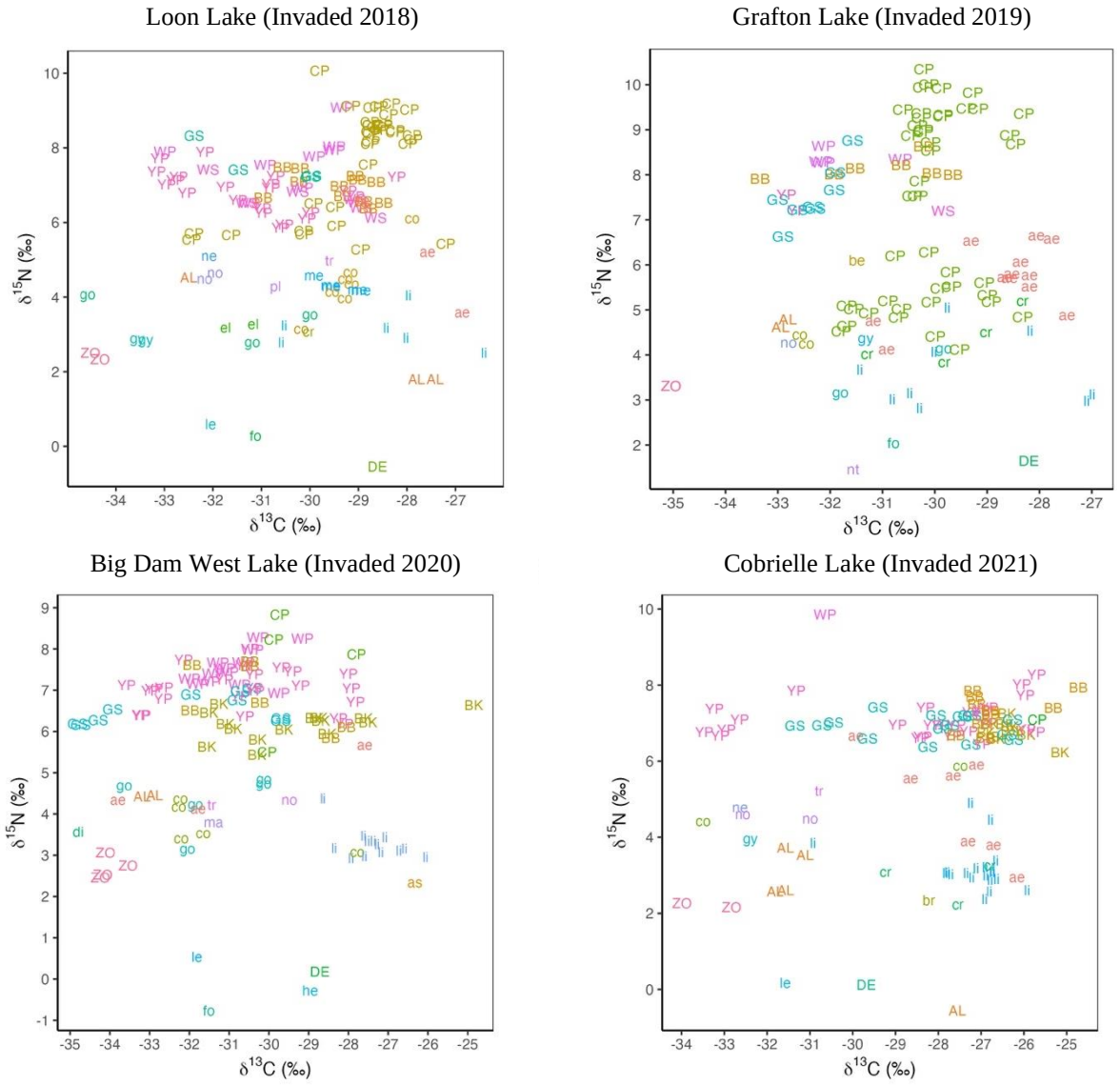

Figure 10: Stable Isotope Bi-Plot of Raw Nitrogen ($\delta^{15}N$) and Carbon ($\delta^{13}C$) Data: This plot illustrates the isotopic composition of nitrogen and carbon in the samples, providing insights into trophic levels and potential food sources. Species codes seen in Table 7.

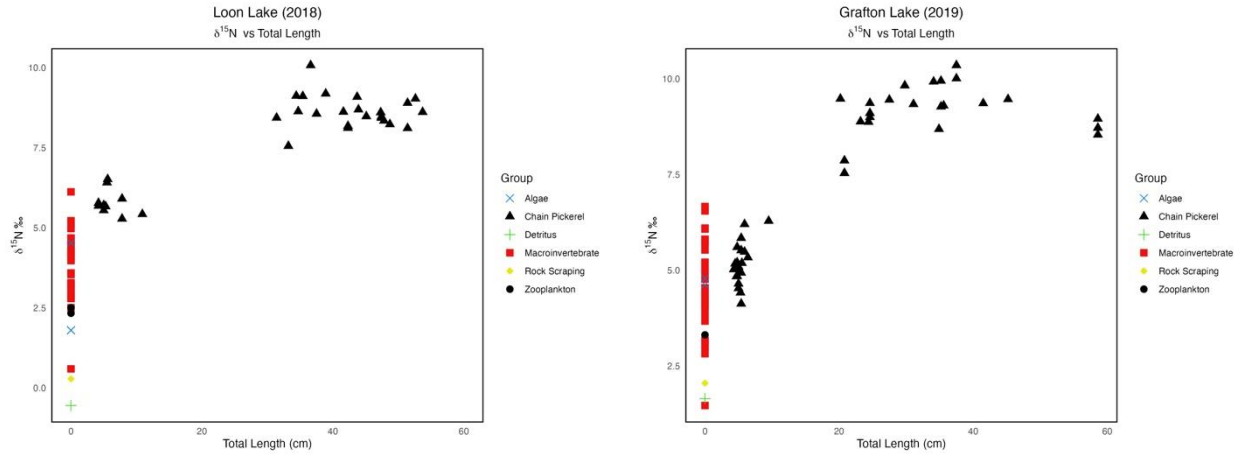

Figure 11: Comparison of E. niger Total Length (cm) δ¹⁵N Stable Isotope values for baseline organisms and for E. niger.

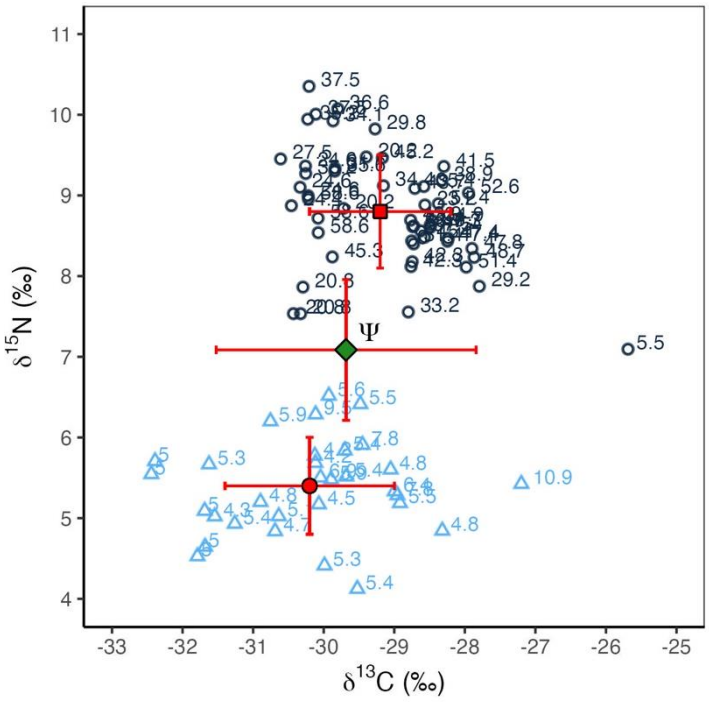

Figure 12: A depiction of E. niger optimally assigned to two distinct groups CP1 and CP2. NbClust was used to determine the optimal number of E. niger clusters in δ-space. NbClust utilizes 30 clustering indices that vote for the optimal number of clusters, the majority of the 30 indices voted for k=2. E. niger were assigned to one of the two clusters using NbClust, clusters were named CP1 and CP2. CP1 had a $\delta^{15}N$ and $\delta^{13}C$ range of 4.1‰ ≤ $\delta^{15}N$ ≤ 6.4‰ and -30.1‰ ≤ $\delta^{13}C$ ≤ -28.9‰ respectively. CP2 had a $\delta^{15}N$ and $\delta^{13}C$ range of 7.1‰ ≤ $\delta^{15}N$ ≤ 10.4‰ and -30.6‰ ≤ $\delta^{13}C$ ≤ -25.7‰ respectively. Average linkage clustering was used to find the centroid (Ψ) between CP1 and CP2 following procedure from 1.5.3 Cluster Analysis. Ψ represent the transition region between clusters CP1 and CP2 and was defined by -29.68 ± 1.84‰, 7.08 ± 0.87‰ in δ–space with standard deviation. CP1 and CP2 centroids were calculated by averaging every individual belonging to each cluster and were found to be (-30.22 ± 1.21‰, 5.38 ± 0.58‰) and (-29.15 ± 0.99‰, 8.82 ± 0.70‰) respectively. The average TL of CP1 and CP2 respectively were 5.69 ± 1.50cm and 36.42 ± 12.63cm with SD. The transition total length (TL) at Ψ was found to be 21.41± 2.24cm using average linkage clustering, 1.5.3 Cluster Analysis. Symbology: circles with no fill (○) = individuals belonging to cluster CP2, triangles with no fill (△) = individuals belonging to cluster CP1, solid square CP2 cluster centroid (■), solid diamond Ψ centroid (◆), solid circle CP1 cluster centroid (●). TL are displayed next to corresponding points in centimeters.

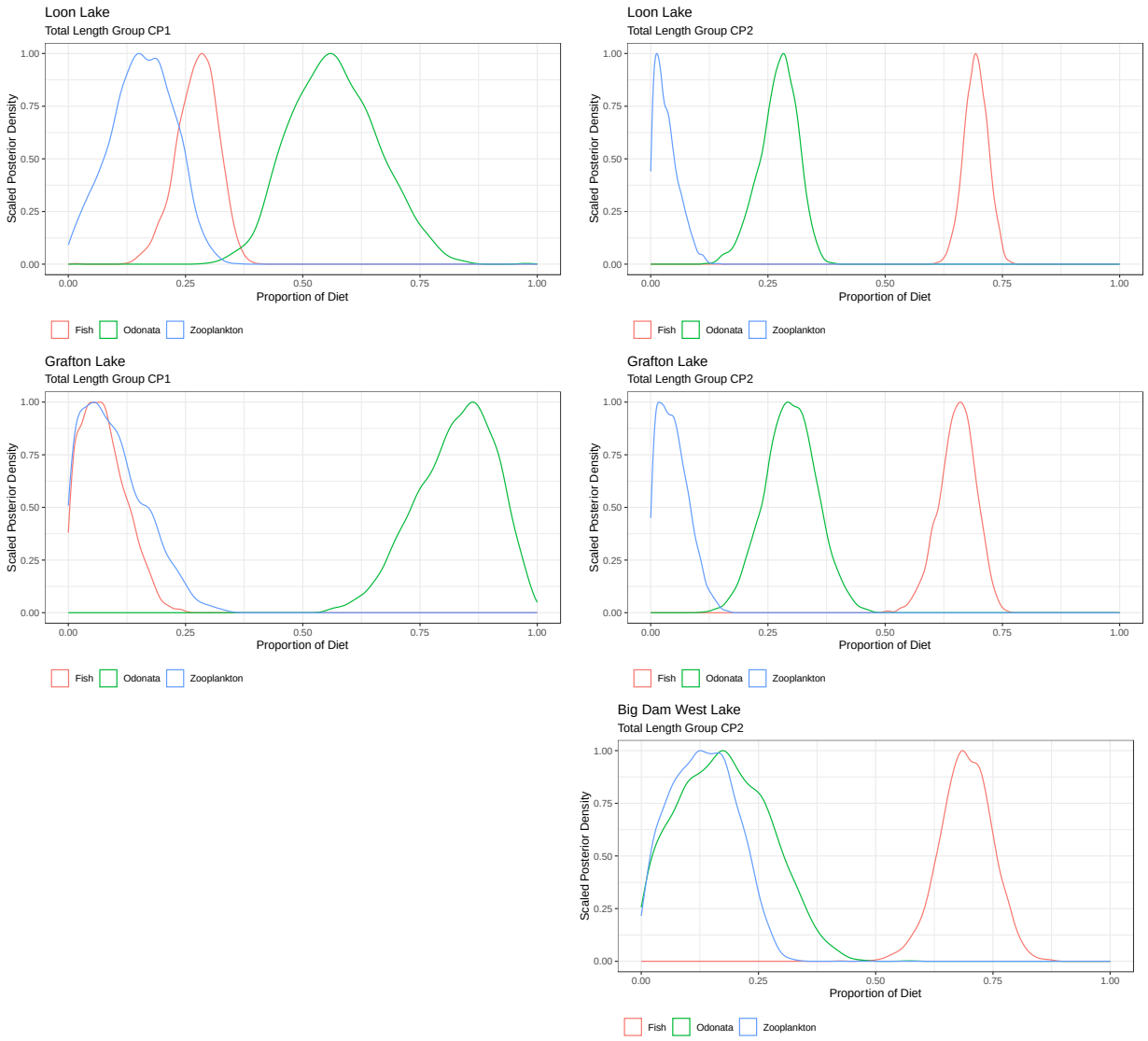

Figure 13: Results of MixSIAR model. Posterior density curves represent the probability distribution of the source's contribution. The peak of the curve indicates the most likely proportion of the source's contribution. The width of the curve gives an indication of the uncertainty around this estimate. Curve with multiple peaks suggest that there are several potential values for the source's contribution.

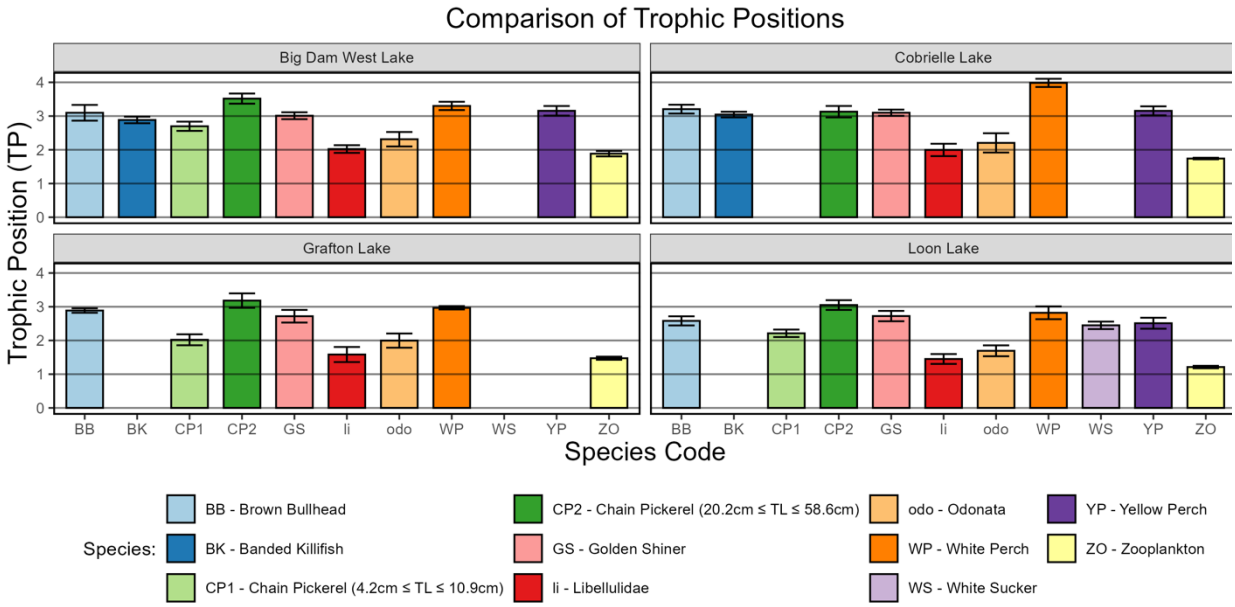

Figure 14: Calculated trophic positions (TP) for species groups for Loon (2018), Grafton (2019), Dam West (2020) and Cobrielle (2021).

2.7 Tables

*Table 2: Mean physical and chemical characteristics of Loon Lake, Grafton lake, Big Dam West Lake, and Cobrielle Lake in Kejimkujik National Park and National Historic Site. (Joseph Kerekes & Peter Schwinghamer, 1973). *Values after Dam Removal in 1995 (Sally O'Grady, 2003).*

Parameter	Unit	Loon Lake	Grafton Lake	Big Dam West Lake	Cobrielle Lake
Latitude	ºN	44.32	44.39	44.46	44.31
Longitude	ºS	-65.19	-65.19	-65.29	-65.23
Water Colour		Brown	Clear	Brown	Clear
pH		5.2179	6.2813	5.2663	5.6208
Turbidity (TDS)	g/L	2.0423	1.5249	0.7714	1.0253
Total Organic Carbon	mg/L	7.9971	5.9183	10.6854	3.2983
Total Nitrogen	mg/L	0.2438	0.27	0.3346	0.1588
Calcium	mg/L	0.5217	0.897	0.6213	0.2633
Surface Area	Ha	73.8	200*	104.7	131.8
Volume	$m^3 \times 10^3$	1470.7	3720*	2593.4	2595.7
Average Depth	m	1.99	2.76*	2.47	1.97
Flushing Rate	$year^{-1}$	418	12.5*	13.1	3.8
Year of first chain pickerel report	year	2018	2019	2020	2021

Table 3: Lakes confirmed to be containing invasive E. niger in KNPNHS. Data provided by Parks Canada (Parks Canada, 2019, 2020).

Location	Easting	Northing	Zone	Date Observed (yyyy-mm-dd)
Mersey River	324592	4910700	20T	2018-06-24
Kejimkujik Lake	322742	4918717	20T	2018-08-22
Peskowesk Brook	326223	4905285	20T	2018-08-23
Loon Lake	325380	4909960	20T	2018-08-20
Rogers Brook	322985	4919381	20T	2018-10-21
Peskowesk Lake	319772	4908636	20T	2018-10-23
Grafton Brook	324455	4916682	20T	2018-07-17
West River	316945	4917583	20T	2019-07-31
Grafton Lake	325932	4916962	20T	2019-10-22
Frozen Ocean Lake	313375	4924805	20T	2020-07-27
Mill Falls	323664	4923024	20T	2020-08-26
Big Dam West Lake	317933	4925814	20T	2020-10-14
Cobrielle Lake	321715	4909149	20T	2021-06-08

Table 4: Summary of fish species sampled for stable isotope analysis by study lake. See Table 9 for the number of chain pickerel in each CP1 and CP2 size class for each lake.

Species	Loon Lake	Grafton Lake	Big Dam West Lake	Cobrielle Lake	Sum
Catostomus commersonii	6	1	0	0	7
Ameiurus nebulosus	10	8	7	8	33
Perca flavescens	20	2	23	25	70
Fundulus diaphanus	0	0	13	9	22
Notemigonus crysoleucas	4	7	9	16	36
Morone americana	7	3	13	1	24
Esox niger	31	38	4	1	74
Sum	78	59	69	60	266

Table 5: Summary of Odonata nymph species sampled for stable isotope analysis by study lake.

Species	Loon Lake	Grafton Lake	Big Dam West Lake	Cobrielle Lake	Sum
Aeshna canadensis	0	5	1	2	8
Aeshna septentrionalis	0	1	1	0	2
Aeshna tuberculifera	0	1	0	0	1
Aeshna umbrosa	2	1	0	2	5
Aeshna verticalis	0	0	0	1	1
Argia fumipennis	1	0	0	0	1
Basiaeschna janata	0	3	1	2	6
Celithemis elisa	2	3	4	5	14
Celithemis eponina	0	0	0	1	1
Celithemis martha	2	0	2	7	11
Argia moesta	1	0	1	1	3
Cordulia shurtleffii	0	0	0	1	1
Dromogomphus spinosus	1	1	1	0	3
Enallagma annexum	1	0	0	0	1
Enallagma aspersum	1	0	0	0	1
Enallagma minusculum	0	1	0	0	1
Enallagma vesperum	2	0	0	0	2
Epitheca cynosura	1	2	1	1	5
Epitheca spinigera	0	2	0	0	2
Hagenius brevistylus	2	1	2	0	5
Helocordulia uhleri	0	0	1	0	1
Ischnura posita	0	0	1	0	1
Ischnura verticalis	0	1	0	1	2
Ladona exusta	1	1	1	0	3
Ladona julia	0	1	2	0	3
Leucorrhinia hudsonica	0	0	0	1	1
Leucorrhinia intacta	0	1	0	0	1
Macromia illinoiensis	0	0	1	0	1
Phanogomphus spicatus	1	0	1	0	2
Plathemis lydia	1	0	0	0	1
Somatochlora elongata	0	0	0	1	1
Stylogomphus albistylus	0	0	1	0	1
Stylurus scudderi	0	0	1	0	1
Sympetrum costiferum	0	1	0	0	1
Sympetrum danae	0	0	1	0	1
Sympetrum internum	0	2	1	3	6
Sympetrum vicinum	0	0	1	0	1
Sum	19	28	26	29	102

Table 6: Summary of zooplankton sampled per study lake for stable isotope analysis. Zooplankton was fractionated into two size classes with 243µm and 53µm Nitex mesh.

Species Name	Size Fraxination (µm)									
	243					53				
	Loon Lake	Grafton Lake	Big Dam West Lake	Cobrielle Lake	Sum	Loon Lake	Grafton Lake	Big Dam West Lake	Cobrielle Lake	Sum
Daphnia (Daphnia) catawba	0	82	7	17	106	0	2	0	0	2
Holopedium gibberum	103	2	114	1	220	1	0	9	0	10
Eubosmina (Eubosmina) longispina	29	0	9	146	184	29	0	0	84	113
Diaphanosoma birgei	3	10	3	2	18	1	0	3	0	4
Bosmina (Bosmina) longirostris	17	0	7	5	29	45	0	11	3	59
Calanoid copepodid	1	30	1	4	36	6	74	32	30	142
Leptodiaptomus minutus	29	6	17	1	53	7	5	4	0	16
Epischura sp. copepodid	3	20	6	8	37	27	64	24	27	142
Calanoid nauplius	0	5	1	7	13	20	24	16	29	89
Cyclopoid nauplius	0	0	1	4	5	0	6	6	21	33
Alona sp.	0	0	0	0	0	0	0	0	1	1
Chydorus sphaericus	0	0	0	0	0	0	0	0	1	1
Daphnia (Daphnia) ambigua	0	0	0	0	0	0	0	0	1	1
Leptodiaptomus siciloides	0	0	0	0	0	0	0	0	1	1
Cyclopoid copepodid	3	1	6	0	10	5	4	46	4	59
Tropocyclops extensus	0	0	0	0	0	0	1	3	1	5
Leptodora kindtii	0	4	3	0	7	0	0	0	0	0
Mesocyclops edax	9	0	2	0	11	0	0	0	0	0
Epischura nordenskioldi	1	4	3	0	8	0	0	1	0	1
Skistodiaptomus pygmaeus	0	0	63	0	63	0	0	3	0	3
Sida crystallina	0	2	0	0	2	0	0	0	0	0
Eubosmina (Neobosmina) tubicen	0	0	0	0	0	16	0	0	0	16
Sum	198	166	243	195	802	157	180	158	203	698

Table 7: List of species codes.

Family	Abbreviation	Family	Abbreviation
Aeshnidae	ae	Gyrinidae	gy
Algae	AL	Helicopsychidae	he
Asellidae	as	Lepido	le
Ictaluridae	BB	Libellulidae	li
Belostomatidae	be	Macromiidae	ma
Fundulidae	BK	Megaloptera	me
Brachycentridae	br	Nepidae	ne
Coenagrionidae	co	Notonectidae	no
Corduliidae	co	Noteridae	nt
Esocidae	CP	Plecoptera	pl
Detritus	DE	Trombidiformes	tr
Dixidae	di	Moronidae	WP
Elimidae	el	Catostomidae	WS
Fontinalaceae	fo	Percidae	YP
Gomphidae	go	Zooplankton	ZO
Cyprinidae	GS		

Table 8: Community Laymen metric calculated using SIBER for each study lake.

Laymen Metric	Big Dam West Lake	Cobrielle Lake	Grafton Lake	Loon Lake
NR	5.61	4.95	5.51	5.92
CR	6.63	4.39	3.55	3.37
TA	21.59	12.25	10.01	14.31
CD	2.17	2.16	2.18	1.97
MNND	1.30	1.00	1.03	0.88
SDNND	1.06	0.89	0.33	0.71

Table 9: Mean $\delta^{13}C$ and $\delta^{15}N$ values for E. niger belonging to cluster CP1 and CP2 separated by study lake.

			Study Lake			
	Cluster	Value	Loon Lake	Grafton Lake	Big dam West Lake	Cobrielle Lake
$\delta^{13}C$	CP1	Mean	-29.08 ± 1.48	-28.98 ± 1.07	-29.1	--
	CP1	n	10	20	1	--
	CP2	Mean	-28.2 ± 0.49	-29.39 ± 0.66	-28.7 ± 1.21	-24.5
	CP2	n	23	24	3	1
$\delta^{15}N$	CP1	Mean	5.79 ± 0.4	5.16 ± 0.56	5.5	--
	CP1	n	10	20	1	--
	CP2	Mean	8.63 ± 0.5	9.12 ± 0.73	8.32 ± 0.49	7.09
	CP2	n	23	24	3	1

Table 10: Mean source values used in MixSIAR model. Lake codes; LL = Loon Lake, GR = Grafton Lake, BD = Big Dam West Lake and CB = Cobrielle Lake.

Lake	Sources	Meand13C	Meand15N	C:N	n
LL	Fish	-30.66 ± 1.74	7.08 ± 0.63	3.34 ± 0.21	52
LL	Zooplankton	-34.42 ± 0.14	2.42 ± 0.12	4.79 ± 0.03	2
LL	Odonata	-28.66 ± 1.63	3.11 ± 0.53	4.31 ± 0.26	6
GR	Fish	-31.95 ± 1.48	7.86 ± 0.55	3.46 ± 0.28	23
GR	Zooplankton	-35.04 ± 0.43	3.31 ± 0.16	4.79 ± 0.09	2
GR	Odonata	-29.45 ± 1.63	3.6 ± 0.79	4.17 ± 0.17	2
BD	Fish	-30.6 ± 1.86	6.86 ± 0.65	3.48 ± 0.38	70
BD	Zooplankton	-34.02 ± 0.32	2.71 ± 0.27	4.67 ± 0.04	4
BD	Odonata	-27.38 ± 0.7	3.28 ± 0.38	4.35 ± 0.2	13
CB	Fish	-26.6 ± 2.14	7.06 ± 0.57	3.66 ± 0.31	65
CB	Zooplankton	-33.41 ± 0.82	2.22 ± 0.08	4.94 ± 0.21	2
CB	Odonata	-27.23 ± 1.03	3.2 ± 0.63	4.35 ± 0.14	18

Table 11: MixSIAR results for CP1 and CP2 consumers and zooplankton, fish, and Odonata. CP1's highest dietary contribution came from with an average proportion of 0.74 ± 0.08. CP2's highest dietary contribution came from fish with an average proportion of 0.74 ± 0.04. SD = standar deviation of souce mean proportion.

TL Group	Lake	Source	Mean	SD
1	Loon Lake	Fish	0.282	0.035
		Odonata	0.536	0.072
		Zooplankton	0.182	0.053
1	Grafton Lake	Fish	0.056	0.044
		Odonata	0.85	0.085
		Zooplankton	0.104	0.073
2	Loon Lake	Fish	0.655	0.038
		Odonata	0.318	0.053
		Zooplankton	0.026	0.03
2	Grafton Lake	Fish	0.696	0.025
		Odonata	0.269	0.039
		Zooplankton	0.036	0.024
2	Big Dam West Lake	Fish	0.684	0.054
		Odonata	0.212	0.088
		Zooplankton	0.104	0.061

Table 12: Calculated trophic positions (TP) for species groups across all four study lakes. The change in TP when comparing Cobrielle and Big Dam West Lakes with Grafton and Loon Lakes shows some native fish species are being displaced due to increased predation pressure. For group codes refer to Table 7. Lake codes; LL = Loon Lake, GR = Grafton Lake, BD = Big Dam West Lake and CB = Cobrielle Lake.

		Study Lake Trophic Positions				
Species	Code	Loon Lake (LO, 2018)	Grafton Lake (GR, 2019)	Big Dam West Lake (BD, 2020)	Cobrielle Lake (CO, 2021)	Change in TP (LO & GR – BD & CO)
Brown Bullhead	BB	2.58 ± 0.14	2.89 ± 0.07	3.10 ± 0.23	3.21 ± 0.13	0.42 ± 0.28
Banded Killifish	BK	--	--	2.88 ± 0.10	3.04 ± 0.08	--
Chain Pickerel	CP1	2.21 ± 0.11	2.02 ± 0.16	2.7 ± 0.14	--	0.58 ± 0.27
Chain Pickerel	CP2	3.05 ± 0.14	3.18 ± 0.21	3.52 ± 0.15	3.13 ± 0.17	0.21 ± 0.34
Native Fish		2.58 ± 0.06	2.86 ± 0.07	3.06 ± 0.05	3.23 ± 0.05	0.43 ± 0.06
Golden Shiner	GS	2.72 ± 0.15	2.72 ± 0.19	3.01 ± 0.10	3.10 ± 0.09	0.33 ± 0.27
Odonata	odo	1.69 ± 0.16	2.00 ± 0.21	2.31 ± 0.21	2.20 ± 0.29	0.41 ± 0.44
White Perch	WP	2.82 ± 0.19	2.97 ± 0.05	3.30 ± 0.12	3.98 ± 0.12	0.75 ± 0.24
White Sucker	WS	2.45 ± 0.11	--	--	--	--
Yellow Perch	YP	2.51 ± 0.16	--	3.16 ± 0.14	3.15 ± 0.13	0.64 ± 0.3
Zooplankton	ZO	1.21 ± 0.04	1.47 ± 0.05	1.88 ± 0.08	1.74 ± 0.02	0.47 ± 0.09

Table 13: CP1 gut content data. Count per species group and proportion are shown.

Prey	Count	Proportion
Banded Killifish	4	0.13
Golden Shiner	3	0.10
Odonata	17	0.55
Yellow Perch	7	0.23
Sum	31	

Table 14: CP2 gut content data. Count per species group, proportion and alpha priors are shown.

Prey	Count	Proportion
Banded Killifish	1	0.04
Brown Bullhead	1	0.04
Golden Shiner	3	0.11
Odonata	4	0.14
White Perch	7	0.25
White Sucker	3	0.11
Yellow Perch	9	0.32
Sum	28	

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Appendix A: Raw Data

Specimen Code	TL (cm)	Common name	Scientific name	d13C	d15N	%C	%N	C/N
HelboreBD001		Caddisfly	*Helicopsyche borealis*	-28.96	-0.27	39.37	5.64	6.98
MicwataCB001		Caddisfly	*Micrasema wataga*	-28.22	2.34	42.78	10.28	Jeff
AL2020Oct06 KNP-001		Algae	n/a	-27.42	1.80	40.56	5.93	6.84
AL2020Oct06 KNP-002		Algae	n/a	-27.80	1.80	43.90	6.00	7.31
AL2020Oct06 KNP-003		Algae	n/a	-32.50	4.52	37.81	6.21	6.09
AL2020Oct06 KNP-007		Algae	n/a	-33.19	4.42	41.13	7.28	5.65
AL2020Oct06 KNP-008		Algae	n/a	-32.87	4.45	40.34	6.90	5.84
AL2020Oct26 KNP-009		Algae	n/a	-32.81	4.79	32.61	5.45	5.99
AL2020Oct26 KNP-010		Algae	n/a	-32.95	4.62	39.65	6.71	5.91
AL2020Oct27 KNP-004		Algae	n/a	-31.81	2.58	24.43	3.80	6.44
AL2020Oct27 KNP-005		Algae	n/a	-31.11	3.53	38.89	6.07	6.41
AL2020Oct27 KNP-005		Algae	n/a	-31.57	3.73	31.43	4.95	6.35
AL2020Oct27 KNP-006		Algae	n/a	-31.57	2.60	24.96	3.65	6.83
RSCB001		Algae	n/a	-27.55	-0.56	39.31	1.52	25.84
CorCorCob001		American emerald	*Cordulia shurtleffii*	-26.80	3.26	45.75	10.60	4.32
SymviciBD001		Autumn Meadowhawk	*Sympetrum vicinum*	-28.64	4.38	46.13	11.00	4.19
EnaaspeLO001		Azure Bluet	*Enallagma aspersum*	-30.18	3.15	49.70	10.80	4.60
AesseptBD004		Azure Darner	*Aeshna septentrionalis*	-33.80	4.35	45.56	11.17	4.08

Specimen Code	TL (cm)	Common name	Scientific name	d13C	d15N	%C	%N	C/N
AesseptGR001		Azure Darner	*Aeshna septentrionalis*	-28.57	5.72	45.86	11.43	4.01
AesseptGR001		Azure Darner	*Aeshna septentrionalis*	-28.66	5.73	45.96	11.53	3.99
AniBueBD005		Backswimmer	*Buenoa*	-29.48	4.35	49.96	10.88	4.59
AniBueCO003		Backswimmer	*Buenoa*	-30.99	4.49	50.16	10.80	4.64
AniBueCO004		Backswimmer	*Buenoa*	-32.57	4.61	49.73	11.12	4.47
AniBueGR001		Backswimmer	*Buenoa*	-32.79	4.28	51.60	10.93	4.72
AniBueLO002		Backswimmer	*Buenoa*	-32.17	4.49	51.41	11.22	4.58
AniBueLO006		Backswimmer	*Buenoa*	-31.96	4.65	51.22	11.00	4.66
BBBD030	16	Banded Killifish	*Ameiurus nebulosus*	-31.98	6.51	46.92	13.93	3.37
BK2020Oct14 KNP-001	7.8	Banded Killifish	*Fundulus diaphanus*	-28.70	6.26	*45.70*	*13.56*	*3.37*
BK2020Oct14 KNP-001	7.8	Banded Killifish	*Fundulus diaphanus*	-28.86	6.33	42.76	12.93	3.31
BK2020Oct14 KNP-002	6.5	Banded Killifish	*Fundulus diaphanus*	-27.62	6.32	47.04	14.14	3.33
BK2020Oct14 KNP-002	6.5	Banded Killifish	*Fundulus diaphanus*	-27.51	6.23	47.23	14.12	3.35
BK2020Oct14 KNP-003	9	Banded Killifish	*Fundulus diaphanus*	-28.75	6.33	47.44	14.07	3.37
BK2020Oct27 KNP-004	6.8	Banded Killifish	*Fundulus diaphanus*	-26.40	7.26	46.66	13.65	3.42
BK2020Oct27 KNP-005	4.3	Banded Killifish	*Fundulus diaphanus*	-26.65	6.58	47.15	12.74	3.70
BK2020Oct27 KNP-006	4.4	Banded Killifish	*Fundulus diaphanus*	-25.15	6.23	45.42	12.76	3.56
BK2020Oct27 KNP-007	5.5	Banded Killifish	*Fundulus diaphanus*	-26.20	6.81	45.85	12.87	3.56
BKBD011	8.2	Banded Killifish	*Fundulus diaphanus*	-31.25	6.67	48.07	12.03	4.00
BKBD012	8.3	Banded Killifish	*Fundulus diaphanus*	-29.62	6.05	50.16	11.92	4.21
BKBD013	4.5	Banded Killifish	*Fundulus diaphanus*	-30.28	5.80	47.82	10.95	4.37

Specimen Code	TL (cm)	Common name	Scientific name	d13C	d15N	%C	%N	C/N
BKBD014	6.5	Banded Killifish	*Fundulus diaphanus*	-30.90	6.07	49.02	12.18	4.02
BKBD015	7.6	Banded Killifish	*Fundulus diaphanus*	-24.85	6.64	42.15	12.27	3.44
BKBD016	7.2	Banded Killifish	*Fundulus diaphanus*	-28.53	5.95	44.90	12.43	3.61
BKBD017	8.1	Banded Killifish	*Fundulus diaphanus*	-31.56	5.63	40.59	10.76	3.77
BKBD018	8.3	Banded Killifish	*Fundulus diaphanus*	-31.51	6.46	42.14	11.02	3.82
BKBD019	7.5	Banded Killifish	*Fundulus diaphanus*	-30.30	5.44	41.77	11.14	3.75
BKBD020	7.9	Banded Killifish	*Fundulus diaphanus*	-31.09	6.18	46.48	12.34	3.77
BKCB009	6.6	Banded Killifish	*Ameiurus nebulosus*	-27.59	6.69	43.46	9.06	4.80
BKCB010	7.4	Banded Killifish	*Ameiurus nebulosus*	-26.91	6.61	49.72	11.28	4.41
BKCB011	7.7	Banded Killifish	*Fundulus diaphanus*	-26.89	6.76	44.62	12.11	3.68
BKCB011	7.7	Banded Killifish	*Fundulus diaphanus*	-26.81	6.97	34.99	9.69	3.61
BKCB012	8	Banded Killifish	*Fundulus diaphanus*	-26.33	6.91	40.62	11.28	3.60
BKCB013	5.9	Banded Killifish	*Fundulus diaphanus*	-26.55	6.64	45.27	12.58	3.60
BKCB014	8.2	Banded Killifish	*Fundulus diaphanus*	-26.44	6.98	41.60	11.94	3.48
BKCB015	7.8	Banded Killifish	*Fundulus diaphanus*	-25.94	6.69	46.87	13.41	3.50
SymdanaBD001		Black Meadowhawk	*Sympetrum danae*	-27.28	3.27	49.20	11.49	4.28
DrospinGR001		Black-Shouldered Spinyleg	*Dromogomphus spinosus*	-31.80	3.17	45.80	9.96	4.60
DrospinLO001		Black-Shouldered Spinyleg	*Dromogomphus spinosus*	-38.78	2.95	50.33	10.85	4.64
DrospinLO002		Black-Shouldered Spinyleg	*Dromogomphus spinosus*	-34.58	4.08	50.35	11.04	4.56

Specimen Code	TL (cm)	Common name	Scientific name	d13C	d15N	%C	%N	C/N
AestubeGR001		Black-Tipped Darner	*Aeshna tuberculifera*	-28.19	5.53	45.28	11.29	4.01
BB2020Oct06KNP-001_Hg	20.6	Brown Bullhead	*Ameiurus nebulosus*	-29.07	7.24	46.82	14.26	3.28
BB2020Oct06KNP-001_SIA	20.6	Brown Bullhead	*Ameiurus nebulosus*	-29.02	7.15	45.38	13.95	3.25
BB2020Oct14KNP-002	14.6	Brown Bullhead	*Ameiurus nebulosus*	-30.48	7.59	45.49	14.08	3.23
BB2020Oct14KNP-003	12.5	Brown Bullhead	*Ameiurus nebulosus*	-30.23	6.69	46.01	13.95	3.30
BB2020Oct14KNP-004	10.4	Brown Bullhead	*Ameiurus nebulosus*	-28.45	5.84	46.79	13.95	3.35
BB2020Oct14KNP-005	12.2	Brown Bullhead	*Ameiurus nebulosus*	-28.05	6.11	46.68	13.30	3.51
BB2020Oct27KNP-006	15.6	Brown Bullhead	*Ameiurus nebulosus*	-27.00	6.99	49.73	13.29	3.74
BBBD031	5.4	Brown Bullhead	*Ameiurus nebulosus*	-30.48	7.70	40.76	10.06	4.05
BBBD032	11.7	Brown Bullhead	*Ameiurus nebulosus*	-31.93	7.61	48.16	13.78	3.49
BBCB001	15.4	Brown Bullhead	*Ameiurus nebulosus*	-26.79	7.33	46.53	12.72	3.66
BBCB002	13	Brown Bullhead	*Ameiurus nebulosus*	-25.31	7.40	47.43	13.23	3.59
BBCB003	14.8	Brown Bullhead	*Ameiurus nebulosus*	-27.15	7.69	48.21	13.29	3.63
BBCB003	14.8	Brown Bullhead	*Ameiurus nebulosus*	-27.20	7.85	*49.14*	*13.39*	*3.67*
BBCB003	14.8	Brown Bullhead	*Ameiurus nebulosus*	-27.10	7.51	49.42	13.72	3.60
BBCB004	16.2	Brown Bullhead	*Ameiurus nebulosus*	-24.73	7.93	44.87	13.07	3.43
BBCB005	13.6	Brown Bullhead	*Ameiurus nebulosus*	-26.78	7.24	49.13	12.77	3.85
BBGR031	18.5	Brown Bullhead	*Ameiurus nebulosus*	-31.92	8.01	46.16	13.27	3.48
BBGR032	16.8	Brown Bullhead	*Ameiurus nebulosus*	-29.65	8.00	33.95	10.04	3.38
BBGR033	15.4	Brown Bullhead	*Ameiurus nebulosus*	-30.22	8.63	47.75	13.57	3.52
BBGR034	15.2	Brown Bullhead	*Ameiurus nebulosus*	-31.52	8.15	44.61	13.25	3.37

Specimen Code	TL (cm)	Common name	Scientific name	d13C	d15N	%C	%N	C/N
BBGR035	16.8	Brown Bullhead	*Ameiurus nebulosus*	-33.34	7.91	45.70	13.26	3.45
BBGR036	17.2	Brown Bullhead	*Ameiurus nebulosus*	-30.06	8.04	47.11	13.99	3.37
BBGR037	16.2	Brown Bullhead	*Ameiurus nebulosus*	-30.64	8.21	46.19	13.18	3.50
BBGR038	13.4	Brown Bullhead	*Ameiurus nebulosus*	-36.64	7.98	42.78	12.83	3.33
BBLO008	6.2	Brown Bullhead	*Ameiurus nebulosus*	-30.96	6.67	45.20	12.18	3.71
BBLO009	17.2	Brown Bullhead	*Ameiurus nebulosus*	-30.56	7.48	46.41	14.01	3.31
BBLO010	18.7	Brown Bullhead	*Ameiurus nebulosus*	-28.63	7.09	45.16	13.74	3.29
BBLO011	21.4	Brown Bullhead	*Ameiurus nebulosus*	-28.48	6.53	46.45	14.17	3.28
BBLO012	20.7	Brown Bullhead	*Ameiurus nebulosus*	-29.32	6.71	*46.79*	*14.08*	*3.32*
BBLO012	20.7	Brown Bullhead	*Ameiurus nebulosus*	-29.39	6.98	54.62	16.89	3.23
BBLO013	20.5	Brown Bullhead	*Ameiurus nebulosus*	-30.22	7.10	*43.59*	*13.18*	*3.31*
BBLO013	20.5	Brown Bullhead	*Ameiurus nebulosus*	-30.21	7.45	45.31	13.99	3.24
BBLO014	20.6	Brown Bullhead	*Ameiurus nebulosus*	-28.80	6.37	*46.77*	*13.93*	*3.36*
BBLO014	20.6	Brown Bullhead	*Ameiurus nebulosus*	-28.89	6.57	46.59	14.34	3.25
BBLO015	17.7	Brown Bullhead	*Ameiurus nebulosus*	-35.82	7.83	*40.28*	*12.22*	*3.30*
BBLO015	17.7	Brown Bullhead	*Ameiurus nebulosus*	-35.90	7.81	46.27	14.17	3.27
RanfuscCB002		Brown Water Scorpion	*Ranatra fusca*	-32.63	4.78	50.25	10.97	4.58
RanfuscLO001		Brown Water Scorpion	*Ranatra fusca*	-32.08	5.11	52.96	10.37	5.11
CelelisBD015		Calico Pennant	*Celithemis elisa*	-26.73	3.12	48.39	11.32	4.27
CelelisBD016		Calico Pennant	*Celithemis elisa*	-27.18	3.08	49.08	11.32	4.34
CelelisCB003		Calico Pennant	*Celithemis elisa*	-26.89	2.99	49.19	11.43	4.30

Specimen Code	TL (cm)	Common name	Scientific name	d13C	d15N	%C	%N	C/N
CelelisCB006		Calico Pennant	*Celithemis elisa*	-25.93	2.61	49.22	10.79	4.56
CelelisCB007		Calico Pennant	*Celithemis elisa*	-26.65	3.38	48.93	11.33	4.32
CelelisCB010		Calico Pennant	*Celithemis elisa*	-27.71	3.03	48.95	11.32	4.33
CelelisGR001		Calico Pennant	*Celithemis elisa*	-31.43	3.67	47.00	11.06	4.25
CelelisGR002		Calico Pennant	*Celithemis elisa*	-26.99	3.13	49.89	11.17	4.47
CelelisGR003		Calico Pennant	*Celithemis elisa*	-27.10	2.98	48.68	11.38	4.28
CelelisLO001		Calico Pennant	*Celithemis elisa*	-28.43	3.17	48.04	11.16	4.30
CelelisLO002		Calico Pennant	*Celithemis elisa*	-30.53	3.24	49.30	10.22	4.82
CelelisCB008		Calico Pennant	*Celithemis elisa*	-26.78	4.46	49.14	10.81	4.54
AescanaBD004		Canada Darner	*Aeshna canadensis*	-27.59	5.68	44.29	11.02	4.02
AescanaCB005		Canada darner	*Aeshna canadensis*	-26.16	2.95	46.90	10.90	4.30
AescanaCB006		Canada darner	*Aeshna canadensis*	-26.71	3.81	45.49	11.19	4.07
AescanaGR001		Canada Darner	*Aeshna canadensis*	-28.53	5.81	46.53	11.27	4.13
AescanaGR002		Canada Darner	*Aeshna canadensis*	-28.06	6.66	46.29	11.34	4.08
AescanaGR003		Canada Darner	*Aeshna canadensis*	-29.31	6.55	45.87	11.43	4.01
AescanaGR004		Canada Darner	*Aeshna canadensis*	-31.18	4.76	50.41	10.32	4.88
AescanaGR009		Canada Darner	*Aeshna canadensis*	-30.92	4.13	52.03	10.33	5.04
CP2020Oct06KNP-001_Hg	42.3	Chain Pickerel	*Esox niger*	-28.75	8.18	47.24	15.04	3.14
CP2020Oct06KNP-001_SIA	42.3	Chain Pickerel	*Esox niger*	-28.76	8.12	44.82	14.23	3.15
CP2020Oct06KNP-002	10.9	Chain Pickerel	*Esox niger*	-27.20	5.43	45.99	14.43	3.19
CP2020Oct06KNP-003	48.7	Chain Pickerel	*Esox niger*	-27.87	8.23	48.16	14.87	3.24

Specimen Code	TL (cm)	Common name	Scientific name	d13C	d15N	%C	%N	C/N
CP2020Oct06 KNP-004	53.7	Chain Pickerel	*Esox niger*	-28.74	8.61	47.75	15.04	3.17
CP2020Oct06 KNP-005	41.6	Chain Pickerel	*Esox niger*	-28.72	8.62	47.02	15.01	3.13
CP2020Oct06 KNP-006	47.8	Chain Pickerel	*Esox niger*	-27.90	8.34	46.38	14.77	3.14
CP2020Oct08 KNP-007	45.1	Chain Pickerel	*Esox niger*	-28.57	8.48	46.63	14.67	3.18
CP2020Oct08 KNP-008	51.4	Chain Pickerel	*Esox niger*	-27.98	8.11	45.51	14.36	3.17
CP2020Oct08 KNP-009	47.3	Chain Pickerel	*Esox niger*	-28.49	8.60	45.40	14.09	3.22
CP2020Oct08 KNP-010	47.4	Chain Pickerel	*Esox niger*	-28.25	8.43	46.59	14.80	3.15
CP2020Oct08 KNP-010	47.4	Chain Pickerel	*Esox niger*	-28.24	8.47	46.78	14.81	3.16
CP2020Oct14 KNP-011	45.3	Chain Pickerel	*Esox niger*	-29.88	8.24	46.70	14.61	3.20
CP2020Oct16 KNP-012	29.2	Chain Pickerel	*Esox niger*	-27.79	7.87	44.80	13.99	3.20
CP2020Oct26 KNP-013	31.1	Chain Pickerel	*Esox niger*	-29.84	9.34	44.38	13.86	3.20
CPBD014	20.2	Chain Pickerel	*Esox niger*	-29.72	8.84	44.78	14.27	3.14
CPBD015	6.9	Chain Pickerel	*Esox niger*	-30.05	5.50	44.33	12.78	3.47
CPCB001	5.5	Chain Pickerel	*Esox niger*	-25.69	7.09	41.47	11.60	3.57
CPGR014	20.2	Chain Pickerel	*Esox niger*	-29.40	9.48	46.09	14.71	3.13
CPGR015	45.2	Chain Pickerel	*Esox niger*	-29.17	9.47	46.52	14.42	3.23
CPGR016	35.2	Chain Pickerel	*Esox niger*	-30.26	9.27	46.94	14.98	3.13
CPGR017	34.9	Chain Pickerel	*Esox niger*	-28.46	8.69	47.04	14.78	3.18
CPGR019	35.2	Chain Pickerel	*Esox niger*	-30.23	9.94	44.52	14.13	3.15
CPGR021	41.5	Chain Pickerel	*Esox niger*	-28.30	9.36	46.02	14.56	3.16
CPGR022	23.2	Chain Pickerel	*Esox niger*	-28.57	8.88	46.54	14.37	3.24

Specimen Code	TL (cm)	Common name	Scientific name	d13C	d15N	%C	%N	C/N
CPGR023	4.8	Chain Pickerel	*Esox niger*	-30.90	5.20	45.52	12.72	3.58
CPGR024	34.1	Chain Pickerel	*Esox niger*	-29.87	9.92	41.12	12.92	3.18
CPGR025	37.5	Chain Pickerel	*Esox niger*	-30.21	10.35	43.80	13.92	3.15
CPGR026	5.1	Chain Pickerel	*Esox niger*	-30.64	5.02	47.41	13.20	3.59
CPGR027	20.8	Chain Pickerel	*Esox niger*	-30.43	7.54	48.17	14.44	3.34
CPGR027	20.8	Chain Pickerel	*Esox niger*	-30.33	7.53	45.31	13.70	3.31
CPGR027	20.8	Chain Pickerel	*Esox niger*	-30.29	7.86	47.37	14.69	3.22
CPGR028	37.5	Chain Pickerel	*Esox niger*	-30.11	10.01	41.79	12.84	3.25
CPGR029	24.4	Chain Pickerel	*Esox niger*	-30.46	8.87	47.02	14.45	3.25
CPGR030	5.9	Chain Pickerel	*Esox niger*	-29.90	5.48	45.32	12.27	3.69
CPGR031	4.8	Chain Pickerel	*Esox niger*	-29.05	5.60	44.73	11.81	3.79
CPGR032	35.6	Chain Pickerel	*Esox niger*	-29.84	9.30	47.09	14.33	3.29
CPGR033	27.5	Chain Pickerel	*Esox niger*	-30.61	9.45	46.57	14.37	3.24
CPGR034	24.6	Chain Pickerel	*Esox niger*	-30.33	9.10	50.09	15.02	3.34
CPGR034	24.6	Chain Pickerel	*Esox niger*	-30.22	9.00	48.27	14.77	3.27
CPGR034	24.6	Chain Pickerel	*Esox niger*	-30.26	9.36	47.66	15.01	3.18
CPGR035	29.8	Chain Pickerel	*Esox niger*	-29.27	9.82	45.45	13.97	3.25
CPGR036	5.4	Chain Pickerel	*Esox niger*	-29.52	4.12	45.21	11.64	3.88
CPGR037	5.3	Chain Pickerel	*Esox niger*	-29.99	4.41	46.16	12.67	3.64
CPGR038	5.4	Chain Pickerel	*Esox niger*	-29.70	5.84	47.53	12.57	3.78
CPGR039	5	Chain Pickerel	*Esox niger*	-31.79	4.53	45.94	12.77	3.60

Specimen Code	TL (cm)	Common name	Scientific name	d13C	d15N	%C	%N	C/N
CPGR040	5	Chain Pickerel	*Esox niger*	-31.68	4.65	47.71	13.31	3.58
CPGR041	9.5	Chain Pickerel	*Esox niger*	-30.11	6.29	44.10	12.04	3.66
CPGR042	5.5	Chain Pickerel	*Esox niger*	-28.92	5.19	48.18	13.18	3.66
CPGR043	4.5	Chain Pickerel	*Esox niger*	-30.07	5.17	45.54	12.85	3.54
CPGR044	5.4	Chain Pickerel	*Esox niger*	-31.26	4.93	47.97	12.26	3.91
CPGR045	5.9	Chain Pickerel	*Esox niger*	-30.75	6.20	46.78	12.79	3.66
CPGR046	5	Chain Pickerel	*Esox niger*	-31.69	5.09	46.00	12.41	3.71
CPGR047	5.4	Chain Pickerel	*Esox niger*	-29.68	5.52	44.80	12.63	3.55
CPGR048	4.8	Chain Pickerel	*Esox niger*	-28.32	4.85	47.30	12.93	3.66
CPGR049	6.4	Chain Pickerel	*Esox niger*	-28.99	5.33	47.01	12.98	3.62
CPGR050	4.7	Chain Pickerel	*Esox niger*	-30.69	4.84	45.54	13.18	3.46
CPGR051	4.3	Chain Pickerel	*Esox niger*	-31.54	5.02	45.72	12.82	3.57
CPGR052	58.6	Chain Pickerel	*Esox niger*	-30.08	8.54	38.32	11.91	3.22
CPGR052	58.6	Chain Pickerel	*Esox niger*	-30.22	8.95	47.03	14.94	3.15
CPGR052	58.6	Chain Pickerel	*Esox niger*	-30.08	8.72	45.70	14.30	3.20
CPLO015	33.2	Chain Pickerel	*Esox niger*	-28.80	7.56	46.04	14.08	3.27
CPLO016	31.4	Chain Pickerel	*Esox niger*	-28.75	8.44	45.83	14.67	3.13
CPLO018	43.7	Chain Pickerel	*Esox niger*	-28.71	9.09	45.00	14.44	3.12
CPLO019	37.5	Chain Pickerel	*Esox niger*	-28.62	8.56	47.57	15.12	3.15
CPLO020	43.9	Chain Pickerel	*Esox niger*	-28.77	8.69	47.11	14.86	3.17
CPLO021	35.4	Chain Pickerel	*Esox niger*	-28.58	9.11	45.45	14.46	3.14

Specimen Code	TL (cm)	Common name	Scientific name	d13C	d15N	%C	%N	C/N
CPLO022	34.4	Chain Pickerel	*Esox niger*	-29.15	9.12	45.90	14.81	3.10
CPLO023		Chain Pickerel	*Esox niger*	-28.73	8.40	37.36	11.80	3.17
CPLO024	34.7	Chain Pickerel	*Esox niger*	-28.49	8.64	45.86	14.56	3.15
CPLO025	38.9	Chain Pickerel	*Esox niger*	-28.32	9.19	47.37	14.88	3.18
CPLO026	52.6	Chain Pickerel	*Esox niger*	-27.95	9.03	45.77	14.23	3.22
CPLO028	5.3	Chain Pickerel	*Esox niger*	-31.63	5.67	46.11	13.07	3.53
CPLO029	5.5	Chain Pickerel	*Esox niger*	-29.48	6.41	47.60	13.35	3.57
CPLO030	7.8	Chain Pickerel	*Esox niger*	-28.96	5.28	47.19	12.59	3.75
CPLO031	51.4	Chain Pickerel	*Esox niger*	-28.37	8.90	45.48	14.72	3.09
CPLO032	5.6	Chain Pickerel	*Esox niger*	-29.93	6.52	47.33	13.56	3.49
CPLO033	5	Chain Pickerel	*Esox niger*	-32.39	5.71	47.40	13.14	3.61
CPLO033	5	Chain Pickerel	*Esox niger*	-32.44	5.55	47.17	12.79	3.69
CPLO034	4.2	Chain Pickerel	*Esox niger*	-30.13	5.77	46.49	13.02	3.57
CPLO034	4.2	Chain Pickerel	*Esox niger*	-30.12	5.69	46.28	13.00	3.56
CPLO035	36.6	Chain Pickerel	*Esox niger*	-29.80	10.08	42.88	14.09	3.04
CPLO036	7.8	Chain Pickerel	*Esox niger*	-29.45	5.91	45.67	13.00	3.51
LadjuliBD002		Chalk-Fronted Corporal	*Ladona julia*	-28.36	3.17	48.60	9.97	4.88
LadjuliGR001		Chalk-Fronted Corporal	*Ladona julia*	-30.48	3.15	42.41	11.06	3.83
SyminteBD001		Cherry-Faced Meadowhawk	*Sympetrum internum*	-27.61	3.48	50.51	11.31	4.46
SyminteCB002		Cherry-Faced	*Sympetrum internum*	-26.74	3.08	44.82	10.69	4.19

Specimen Code	TL (cm)	Common name	Scientific name	d13C	d15N	%C	%N	C/N
		Meadowhawk						
SyminteCB010		Cherry-Faced Meadowhawk	*Sympetrum internum*	-27.35	3.06	48.62	11.56	4.21
SyminteCB010		Cherry-Faced Meadowhawk	*Sympetrum internum*	-27.11	3.19	49.24	11.54	4.27
SyminteCB011		Cherry-Faced Meadowhawk	*Sympetrum internum*	-26.79	2.88	49.57	11.24	4.41
SyminteGR001		Cherry-Faced Meadowhawk	*Sympetrum internum*	-29.76	5.05	44.97	10.89	4.13
SyminteGR002		Cherry-Faced Meadowhawk	*Sympetrum internum*	-30.30	2.82	43.40	10.46	4.15
NotHydGR001		Coleoptera	*Hydrocanthus*	-31.57	1.46	45.71	9.79	4.67
EpicynoBD001		Common Baskettail	*Epitheca cynosura*	-27.78	3.09	45.36	10.05	4.51
EpicynoCB001		Common Baskettail	*Epitheca cynosura*	-29.23	3.09	45.36	10.84	4.18
EpicynoGR001		Common Baskettail	*Epitheca cynosura*	-29.82	3.84	46.10	10.19	4.53
EpicynoGR003		Common Baskettail	*Epitheca cynosura*	-31.29	4.03	45.03	10.54	4.27
EpicynoLO001		Common Baskettail	*Epitheca cynosura*	-30.03	3.08	45.21	10.21	4.43
PlalydiLO001		Common Whitetail	*Plathemis lydia*	-28.02	2.90	44.34	10.40	4.26
DTBD001		Detritus	n/a	-28.72	0.18	51.76	1.18	43.69
DTCB001		Detritus	n/a	-29.68	0.12	48.79	0.95	51.34
DTGR001		Detritus	n/a	-28.20	1.65	43.00	0.85	50.44
DTLO001		Detritus	n/a	-28.61	-0.55	52.33	0.98	53.40

Specimen Code	TL (cm)	Common name	Scientific name	d13C	d15N	%C	%N	C/N
LeuintaGR001		Dot-Tailed Whiteface	*Leucorrhinia intacta*	-30.01	4.07	46.73	11.08	4.22
HagbrevBD001		Dragonhunter	*Hagenius brevistylus*	-31.85	4.25	45.96	11.20	4.11
HagbrevBD002		Dragonhunter	*Hagenius brevistylus*	-32.05	3.16	47.91	11.17	4.29
HagbrevGR001		Dragonhunter	*Hagenius brevistylus*	-29.82	4.15	49.84	10.60	4.70
HagbrevLO001		Dragonhunter	*Hagenius brevistylus*	-29.99	3.54	53.06	9.49	5.59
HagbrevLO002		Dragonhunter	*Hagenius brevistylus*	-31.19	2.80	52.02	10.10	5.15
PhaspicBD001		Dusky Clubtail	*Phanogomphus spicatus*	-30.12	4.85	45.27	10.97	4.13
PhaspicLO010		Dusky Clubtail	*Phanogomphus spicatus*	-38.69	3.24	48.14	10.62	4.53
IscvertCB001		Eastern Forktail	*Ischnura verticalis*	-33.49	4.43	49.85	10.96	4.55
IscvertGR001		Eastern Forktail	*Ischnura verticalis*	-32.45	4.26	46.68	10.22	4.57
StyalbiBD001		Eastern Least Clubtail	*Stylogomphus albistylus*	-33.63	4.70	45.87	11.30	4.06
IscposiBD001		Fragile Forktail	*Ischnura posita*	-32.20	3.42	48.43	10.27	4.71
TriphryBD001		Giant Casemakers	n/a	-26.42	1.71	48.40	9.00	5.38
LetamerGR001		Giant Water Bug	*Lethocerus americanus*	-31.49	6.10	52.75	10.41	5.07
GS2020Oct06KNP-001_Hg	13.1	Golden Shiner	*Notemigonus crysoleucas*	-29.97	7.25	45.84	13.99	3.28
GS2020Oct06KNP-001_SIA	13.1	Golden Shiner	*Notemigonus crysoleucas*	-29.99	7.23	46.68	14.33	3.26
GS2020Oct14KNP-002	10.5	Golden Shiner	*Notemigonus crysoleucas*	-31.96	6.91	48.27	14.17	3.41

Specimen Code	TL (cm)	Common name	Scientific name	d13C	d15N	%C	%N	C/N
GS2020Oct27 KNP-003	6.6	Golden Shiner	*Notemigonus crysoleucas*	-27.31	7.21	47.38	14.36	3.30
GS2020Oct27 KNP-004	6.8	Golden Shiner	*Notemigonus crysoleucas*	-26.27	7.09	46.35	13.82	3.35
GS2020Oct27 KNP-005	6.4	Golden Shiner	*Notemigonus crysoleucas*	-27.74	6.95	47.42	14.03	3.38
GS2020Oct27 KNP-006	6.2	Golden Shiner	*Notemigonus crysoleucas*	-28.05	7.21	48.55	13.90	3.49
GSBD015	9.2	Golden Shiner	*Notemigonus crysoleucas*	-33.93	6.54	52.43	11.75	4.46
GSBD016	7.4	Golden Shiner	*Notemigonus crysoleucas*	-34.29	6.29	49.17	12.04	4.08
GSBD017	6.4	Golden Shiner	*Notemigonus crysoleucas*	-34.81	6.19	51.02	10.88	4.69
GSBD018	6.6	Golden Shiner	*Notemigonus crysoleucas*	-34.73	6.16	49.76	11.38	4.37
GSBD019	11.4	Golden Shiner	*Notemigonus crysoleucas*	-30.80	6.77	47.53	14.29	3.33
GSBD020	11.8	Golden Shiner	*Notemigonus crysoleucas*	-30.71	6.98	40.03	12.12	3.30
GSBD021	7.3	Golden Shiner	*Notemigonus crysoleucas*	-29.70	6.27	42.14	11.74	3.59
GSBD021	7.3	Golden Shiner	*Notemigonus crysoleucas*	-29.69	6.32	44.09	12.27	3.59

Specimen Code	TL (cm)	Common name	Scientific name	d13C	d15N	%C	%N	C/N
GSBD022	10.9	Golden Shiner	*Notemigonus crysoleucas*	-30.42	7.05	46.74	14.25	3.28
GSCB009	5.8	Golden Shiner	*Notemigonus crysoleucas*	-30.72	6.95	44.55	11.22	3.97
GSCB010	4.5	Golden Shiner	*Notemigonus crysoleucas*	-30.44	7.02	47.62	11.98	3.98
GSCB011	4.6	Golden Shiner	*Notemigonus crysoleucas*	-31.35	6.94	48.84	10.97	4.45
GSCB012	3.8	Golden Shiner	*Notemigonus crysoleucas*	-29.41	7.42	45.50	10.97	4.15
GSCB013	6.1	Golden Shiner	*Notemigonus crysoleucas*	-29.65	6.60	46.29	11.92	3.89
GSCB014	5.2	Golden Shiner	*Notemigonus crysoleucas*	-27.90	6.86	43.80	11.60	3.78
GSCB015	7.4	Golden Shiner	*Notemigonus crysoleucas*	-28.25	6.38	42.21	12.07	3.50
GSCB016	9.7	Golden Shiner	*Notemigonus crysoleucas*	-27.22	7.20	46.41	14.00	3.32
GSCB017	9.6	Golden Shiner	*Notemigonus crysoleucas*	-26.40	6.71	47.00	14.28	3.29
GSCB018	9.4	Golden Shiner	*Notemigonus crysoleucas*	-27.44	7.18	45.45	13.71	3.31
GSCB019	9.1	Golden Shiner	*Notemigonus crysoleucas*	-27.24	6.45	44.86	11.65	3.85

Specimen Code	TL (cm)	Common name	Scientific name	d13C	d15N	%C	%N	C/N
GSCB020	8.4	Golden Shiner	*Notemigonus crysoleucas*	-26.24	6.57	37.95	10.35	3.67
GSGR001	16.7	Golden Shiner	*Notemigonus crysoleucas*	-31.57	8.76	47.50	14.78	3.21
GSGR002	8.1	Golden Shiner	*Notemigonus crysoleucas*	-33.01	7.45	50.50	12.06	4.19
GSGR003	7.9	Golden Shiner	*Notemigonus crysoleucas*	-32.33	7.29	48.85	12.58	3.88
GSGR004	8.3	Golden Shiner	*Notemigonus crysoleucas*	-32.29	7.25	38.20	10.96	3.48
GSGR004	8.3	Golden Shiner	*Notemigonus crysoleucas*	-32.63	7.23	40.97	11.20	3.66
GSGR005	9.2	Golden Shiner	*Notemigonus crysoleucas*	-32.90	6.63	41.62	12.28	3.39
GSGR006	11	Golden Shiner	*Notemigonus crysoleucas*	-31.90	8.06	46.64	14.27	3.27
GSGR007	10.1	Golden Shiner	*Notemigonus crysoleucas*	-31.92	7.67	45.98	14.37	3.20
GSLO007	10.1	Golden Shiner	*Notemigonus crysoleucas*	-31.47	7.42	45.96	14.11	3.26
GSLO008	14.4	Golden Shiner	*Notemigonus crysoleucas*	-32.39	8.33	36.02	11.02	3.27
RSLO004		Greater Water Moss	*Fontinalis antipyretica*	-31.12	0.28	33.13	1.94	17.08

Specimen Code	TL (cm)	Common name	Scientific name	d13C	d15N	%C	%N	C/N
RSBD001		Greater Water-Moss	*Fontinalis antipyretica*	-31.51	-0.76	35.43	1.76	20.18
RSGR001		Greater Water-Moss	*Fontinalis antipyretica*	-30.79	2.05	24.86	1.06	23.48
AesvertCB004		Green-Striped Darner	*Aeshna verticalis*	-27.30	3.90	45.90	11.03	4.16
CelelisBD011		Halloween Pennant	*Celithemis elisa*	-26.06	2.96	47.77	10.78	4.43
CelelisBD012		Halloween Pennant	*Celithemis elisa*	-27.38	3.35	49.08	11.31	4.34
CelelisBD012		Halloween Pennant	*Celithemis elisa*	-27.60	2.98	47.89	10.99	4.36
CeleponCB001		Halloween Pennant	*Celithemis eponina*	-26.92	2.37	45.36	10.63	4.27
LeuhudsCB001		Hudsonian Whiteface	*Leucorrhinia hudsonica*	-26.81	2.57	44.22	10.77	4.10
CaecidBD002		Isopoda	*Caecidotea*	-26.33	2.35	31.40	6.47	4.85
EnaminuGR001		Little Bluet	*Enallagma minusculum*	-32.58	4.46	48.83	10.54	4.63
CelmartBD008		Martha's Pennant	*Celithemis martha*	-27.53	3.35	45.42	10.77	4.22
CelmartBD009		Martha's Pennant	*Celithemis martha*	-26.59	3.16	48.20	10.89	4.43
CelmartCB004		Martha's Pennant	*Celithemis martha*	-27.79	3.09	46.63	10.62	4.39
CelmartCB006		Martha's Pennant	*Celithemis martha*	-27.22	2.94	47.74	10.62	4.49
CelmartCB008		Martha's Pennant	*Celithemis martha*	-27.83	3.05	46.60	10.46	4.46
CelmartCB009		Martha's Pennant	*Celithemis martha*	-26.91	3.22	49.66	11.28	4.40
CelmartCB009		Martha's Pennant	*Celithemis martha*	-26.74	3.11	49.44	11.08	4.46
CelmartCob001		Martha's Pennant	*Celithemis martha*	-30.92	3.85	45.89	10.84	4.23

Specimen Code	TL (cm)	Common name	Scientific name	d13C	d15N	%C	%N	C/N
CelmartCob002		Martha's Pennant	*Celithemis martha*	-26.64	2.90	43.91	10.72	4.09
CelmartLO006		Martha's Pennant	*Celithemis martha*	-26.41	2.50	44.14	10.58	4.17
CelmartCB010		Martha's Pennant	*Celithemis martha*	-27.25	4.91	49.80	11.34	4.39
CelmartLO008		Martha's Pennant	*Celithemis martha*	-27.97	4.04	49.73	12.05	4.13
DixidaBD001		Meniscus Midges	n/a	-34.79	3.57	47.46	11.06	4.29
LepidoBD001		Moth or Buterfly	n/a	-31.81	0.55	45.49	8.62	5.28
LepidoCB003		Moth or Buterfly	n/a	-31.57	0.17	40.55	8.84	4.59
LepidoLO002		Moth or Buterfly	n/a	-32.05	0.60	41.35	8.10	5.10
EnaanneLO001		Northern Bluet	*Enallagma annexum*	-29.54	4.15	49.59	11.69	4.24
CoeArgBD003		Powdered Dancer	*Argia moesta*	-31.65	3.54	46.30	11.36	4.07
CoeArgCB001		Powdered Dancer	*Argia moesta*	-27.48	5.88	45.59	10.94	4.17
CoeArgLO002		Powdered Dancer	*Argia moesta*	-27.89	6.12	45.75	11.51	3.97
SymcostGR001		Saffron-Winged Meadowhawk	*Sympetrum costiferum*	-28.18	4.54	42.75	10.59	4.04
AesumbrGR001		Shadow Darner	*Aeshna umbrosa*	-27.47	4.89	45.44	10.77	4.22
AesumbrLO002		Shadow Darner	*Aeshna umbrosa*	-26.86	3.59	45.47	10.84	4.19
AesumbrLO003		Shadow Darner	*Aeshna umbrosa*	-27.58	5.22	49.52	10.72	4.62
AesumbrCB001		Shadow Darner	*Aeshna umbrosa*	-27.65	5.63	48.72	11.55	4.22
AesumbrCB002		Shadow Darner	*Aeshna umbrosa*	-28.65	5.55	49.69	11.93	4.17
SomelonCB001		Ski-Tipped Emerald	*Somatochlora elongata*	-27.54	2.24	46.04	10.26	4.49
EpispinGR001		Spiny Baskettail	*Epitheca spinigera*	-28.32	5.21	47.95	10.04	4.78

Specimen Code	TL (cm)	Common name	Scientific name	d13C	d15N	%C	%N	C/N
EpispinGR002		Spiny Baskettail	*Epitheca spinigera*	-29.02	4.51	47.14	10.29	4.58
BasjanaBD001		Springtime Darner	*Basiaeschna janata*	-31.78	4.14	48.36	10.84	4.46
BasjanaGR001		Springtime Darner	*Basiaeschna janata*	-28.36	6.07	45.57	11.89	3.83
BasjanaGR002		Springtime Darner	*Basiaeschna janata*	-27.77	6.59	45.62	11.66	3.91
BasjanaGR003		Springtime Darner	*Basiaeschna janata*	-28.19	5.78	46.00	11.76	3.91
BasjanaCB004		Springtime Darner	*Basiaeschna janata*	-29.92	6.68	46.33	11.94	3.88
BasjanaCB005		Springtime Darner	*Basiaeschna janata*	-27.10	5.91	44.68	11.39	3.92
MacilliBD001		Swift River Cruiser	*Macromia illinoiensis*	-31.39	3.81	43.41	11.10	3.91
HeluhleBD001		Uhler's Sundragon	*Helocordulia uhleri*	-32.22	4.37	44.53	11.08	4.02
HeluhleBD001		Uhler's Sundragon	*Helocordulia uhleri*	-32.26	4.19	44.72	11.16	4.01
ArgfumiLO001		Variable Dancer	*Argia fumipennis*	-29.16	4.68	48.17	10.54	4.57
EnavespLO001		Vesper Bluet	*Enallagma vesperum*	-29.28	3.98	47.87	11.50	4.16
EnavespLO002		Vesper Bluet	*Enallagma vesperum*	-29.27	4.49	47.97	11.42	4.20
EnavespLO002		Vesper Bluet	*Enallagma vesperum*	-29.14	4.36	49.11	11.86	4.14
TroHydrBD001		Water Mite	n/a	-31.43	4.22	59.47	7.32	8.12
TroHydrLO001		Water Mite	n/a	-29.60	4.98	50.23	10.53	4.77
GyrDinCO004		Whirligig Beetle	*Dineutus*	-32.39	3.97	56.64	8.20	6.91
GyrDinGR003		Whirligig Beetle	*Dineutus*	-31.32	4.38	55.18	8.27	6.67
GyrDinLO001		Whirligig Beetle	*Dineutus*	-33.56	2.89	54.00	8.41	6.42
GyrDinLO002		Whirligig Beetle	*Dineutus*	-33.38	2.86	55.17	8.00	6.90

Specimen Code	TL (cm)	Common name	Scientific name	d13C	d15N	%C	%N	C/N
LadexusBD001		White Corporal	*Ladona exusta*	-27.09	3.43	49.06	11.43	4.29
LadexusGR002		White Corporal	*Ladona exusta*	-30.81	3.02	43.81	10.52	4.16
LadexusLO001		White Corporal	*Ladona exusta*	-30.59	2.79	45.87	11.02	4.16
LadjuliBD001		White Corporal	*Ladona julia*	-27.93	2.93	47.05	11.70	4.02
WP2020Oct06 KNP-001	32.3	White Perch	*Morone americana*	-29.35	9.08	47.91	14.95	3.20
WP2020Oct14 KNP-002	19.7	White Perch	*Morone americana*	-29.75	6.93	43.44	14.10	3.08
WP2020Oct14 KNP-003	17.4	White Perch	*Morone americana*	-31.28	7.67	47.91	14.92	3.21
WP2020Oct15 KNP-004	21.1	White Perch	*Morone americana*	-30.27	8.28	47.57	14.86	3.20
WP2020Oct15 KNP-005	22.5	White Perch	*Morone americana*	-29.16	8.25	46.36	14.46	3.21
WP2020Oct15 KNP-006	20	White Perch	*Morone americana*	-31.12	7.53	46.79	14.78	3.17
WP2020Oct15 KNP-007	18.8	White Perch	*Morone americana*	-31.99	7.28	47.59	14.73	3.23
WPBD001	15.4	White Perch	*Morone americana*	-30.41	7.98	45.67	14.20	3.22
WPBD002	12.4	White Perch	*Morone americana*	-31.04	7.45	48.81	14.75	3.31
WPBD003	7.8	White Perch	*Morone americana*	-30.74	7.13	46.26	14.12	3.28
WPBD004	15.6	White Perch	*Morone americana*	-31.79	7.15	45.81	14.01	3.27
WPBD005	13.5	White Perch	*Morone americana*	-31.40	7.41	48.01	14.74	3.26
WPBD006	17.3	White Perch	*Morone americana*	-30.64	7.68	47.46	14.60	3.25
WPBD010	14.8	White Perch	*Morone americana*	-31.50	7.21	47.12	14.37	3.28
WPCB001	27.6	White Perch	*Morone americana*	-30.65	9.86	44.26	13.22	3.35
WPGR001	16.1	White Perch	*Morone americana*	-30.66	8.36	45.61	14.00	3.26
WPGR002	11.7	White Perch	*Morone americana*	-32.13	8.27	45.87	14.25	3.22

Specimen Code	TL (cm)	Common name	Scientific name	d13C	d15N	%C	%N	C/N
WPGR002	11.7	White Perch	*Morone americana*	-32.17	8.30	45.29	14.06	3.22
WPGR003	12.7	White Perch	*Morone americana*	-32.13	8.64	44.53	13.58	3.28
WPLO001	26.4	White Perch	*Morone americana*	-29.50	7.95	47.59	14.92	3.19
WPLO002	26.1	White Perch	*Morone americana*	-30.16	6.94	45.09	14.33	3.15
WPLO003	25.2	White Perch	*Morone americana*	-30.92	7.54	44.28	14.06	3.15
WPLO004	25.7	White Perch	*Morone americana*	-29.91	7.78	45.91	14.59	3.15
WPLO005	24.5	White Perch	*Morone americana*	-29.50	8.04	46.00	14.37	3.20
WPLO006	24.4	White Perch	*Morone americana*	-32.99	7.90	46.90	14.69	3.19
WS2020Oct06 KNP-001	34.8	White Sucker	*Catostomus commersonii*	-30.25	6.83	47.87	14.97	3.20
WS2020Oct06 KNP-001_Hg	34.8	White Sucker	*Catostomus commersonii*	-31.25	6.53	47.57	14.96	3.18
WS2020Oct06 KNP-001_SIA		White Sucker	*Catostomus commersonii*	-31.30	6.53	47.39	14.86	3.19
WS2020Oct08 KNP-003	36.4	White Sucker	*Catostomus commersonii*	-29.08	6.63	45.19	14.28	3.16
WS2020Oct08 KNP-003	36.4	White Sucker	*Catostomus commersonii*	-29.00	6.54	44.74	13.92	3.21
WS2020Oct08 KNP-003	36.4	White Sucker	*Catostomus commersonii*	-28.95	6.40	38.89	12.17	3.20
WSGR005	34.3	White Sucker	*Catostomus commersonii*	-29.84	7.20	44.34	13.73	3.23
WSLO006	35.2	White Sucker	*Catostomus*	-28.65	6.13	44.89	14.09	3.19

Specimen Code	TL (cm)	Common name	Scientific name	d13C	d15N	%C	%N	C/N
			commersonii					
WSLO007	31.4	White Sucker	*Catostomus commersonii*	-32.10	7.42	45.68	14.38	3.18
BB2020Oct27 KNP-007	7.2	Yellow Perch	*Perca flavescens*	-26.98	6.45	47.78	12.94	3.69
YP2020Oct06 KNP-001_Hg	7.4	Yellow Perch	*Perca flavescens*	-32.71	7.22	45.57	13.43	3.39
YP2020Oct06 KNP-001_SIA	7.4	Yellow Perch	*Perca flavescens*	-33.16	7.36	46.60	13.48	3.46
YP2020Oct06 KNP-002_Hg	6.2	Yellow Perch	*Perca flavescens*	-32.16	7.89	45.73	13.72	3.33
YP2020Oct06 KNP-003_Hg	7.9	Yellow Perch	*Perca flavescens*	-29.22	6.85	46.56	14.13	3.30
YP2020Oct06 KNP-003_SIA	7.9	Yellow Perch	*Perca flavescens*	-29.06	6.69	45.76	14.02	3.26
YP2020Oct06 KNP-004_Hg	7.1	Yellow Perch	*Perca flavescens*	-32.77	7.14	44.51	13.41	3.32
YP2020Oct06 KNP-004_SIA	7.1	Yellow Perch	*Perca flavescens*	-32.95	7.02	45.78	13.43	3.41
YP2020Oct06 KNP-005	11.3	Yellow Perch	*Perca flavescens*	-28.21	7.23	47.67	14.36	3.32
YP2020Oct14 KNP-006	16.1	Yellow Perch	*Perca flavescens*	-29.40	7.46	44.75	14.01	3.19
YP2020Oct14 KNP-007	18.6	Yellow Perch	*Perca flavescens*	-30.47	8.01	46.26	14.75	3.14
YP2020Oct14 KNP-008	18.4	Yellow Perch	*Perca flavescens*	-31.13	7.26	46.91	15.02	3.12
YP2020Oct14 KNP-009	12.7	Yellow Perch	*Perca flavescens*	-30.60	6.37	46.89	14.95	3.14
YP2020Oct14 KNP-010	8.7	Yellow Perch	*Perca flavescens*	-28.25	6.32	46.45	14.40	3.22
YP2020Oct14 KNP-011	10.8	Yellow Perch	*Perca flavescens*	-28.00	7.40	46.93	14.71	3.19
YP2020Oct16 KNP-012	6.8	Yellow Perch	*Perca flavescens*	-33.22	6.40	46.36	14.05	3.30
YP2020Oct16 KNP-012	6.8	Yellow Perch	*Perca flavescens*	-33.20	6.40	45.79	13.92	3.29
YP2020Oct16 KNP-013	6.8	Yellow Perch	*Perca flavescens*	-33.58	7.13	46.20	13.65	3.39
YP2020Oct27 KNP-014	15.6	Yellow Perch	*Perca flavescens*	-25.96	7.74	47.88	14.50	3.30

Specimen Code	TL (cm)	Common name	Scientific name	d13C	d15N	%C	%N	C/N
YP2020Oct27 KNP-015	14	Yellow Perch	*Perca flavescens*	-25.70	8.28	47.51	14.81	3.21
YP2020Oct27 KNP-016	7.2	Yellow Perch	*Perca flavescens*	-27.58	6.96	47.17	14.49	3.26
YP2020Oct28 KNP-017	9.7	Yellow Perch	*Perca flavescens*	-26.84	7.00	45.40	14.31	3.17
YPBD033	11.8	Yellow Perch	*Perca flavescens*	-27.93	7.05	46.76	14.60	3.20
YPBD034	9.9	Yellow Perch	*Perca flavescens*	-30.38	7.06	45.37	14.20	3.20
YPBD035	8.9	Yellow Perch	*Perca flavescens*	-30.34	6.99	45.76	13.89	3.29
YPBD036	10.4	Yellow Perch	*Perca flavescens*	-29.20	7.12	45.85	14.32	3.20
YPBD037	4.4	Yellow Perch	*Perca flavescens*	-32.88	7.03	46.81	12.52	3.74
YPBD038	4.5	Yellow Perch	*Perca flavescens*	-32.65	6.79	45.46	12.53	3.63
YPBD039	8.6	Yellow Perch	*Perca flavescens*	-28.10	6.19	47.41	14.49	3.27
YPBD040	9.6	Yellow Perch	*Perca flavescens*	-27.82	6.72	47.84	14.77	3.24
YPBD041	10.1	Yellow Perch	*Perca flavescens*	-30.36	7.39	46.88	14.16	3.31
YPBD042	6.3	Yellow Perch	*Perca flavescens*	-32.98	6.99	52.23	11.60	4.50
YPBD043	11.7	Yellow Perch	*Perca flavescens*	-30.73	6.86	47.86	14.52	3.30
YPBD044	12.1	Yellow Perch	*Perca flavescens*	-29.68	7.55	48.15	14.77	3.26
YPBD045	18.6	Yellow Perch	*Perca flavescens*	-30.61	7.61	45.63	14.27	3.20
YPBD045	18.6	Yellow Perch	*Perca flavescens*	-30.53	7.59	45.51	14.33	3.18
YPBD046	6.9	Yellow Perch	*Perca flavescens*	-32.64	7.07	47.21	12.34	3.82
YPBD047		Yellow Perch	*Perca flavescens*	-32.14	7.74	44.63	13.65	3.27
YPCB030	7.6	Yellow Perch	*Perca flavescens*	-27.23	7.29	44.40	13.04	3.40
YPCB031	6.5	Yellow Perch	*Perca flavescens*	-26.99	7.35	43.34	12.66	3.42

Specimen Code	TL (cm)	Common name	Scientific name	d13C	d15N	%C	%N	C/N
YPCB032	13.6	Yellow Perch	*Perca flavescens*	-26.05	8.02	47.39	14.67	3.23
YPCB037	8.6	Yellow Perch	*Perca flavescens*	-25.94	6.85	41.31	11.23	3.68
YPCB038	8.1	Yellow Perch	*Perca flavescens*	-27.67	6.66	37.76	10.31	3.66
YPCB039	8.2	Yellow Perch	*Perca flavescens*	-31.32	7.85	38.16	9.11	4.19
YPCB040	4.6	Yellow Perch	*Perca flavescens*	-33.10	6.67	43.64	11.07	3.94
YPCB041	4.1	Yellow Perch	*Perca flavescens*	-32.64	7.10	44.99	12.24	3.68
YPCB042	3.7	Yellow Perch	*Perca flavescens*	-32.95	6.83	43.24	11.46	3.77
YPCB043	4.5	Yellow Perch	*Perca flavescens*	-33.47	6.77	46.30	11.90	3.89
YPCB044	N/A	Yellow Perch	*Perca flavescens*	-27.29	6.79	45.88	11.83	3.88
YPCB045	4.4	Yellow Perch	*Perca flavescens*	-25.72	6.76	43.74	12.03	3.64
YPCB046	N/A	Yellow Perch	*Perca flavescens*	-33.23	7.37	42.77	10.98	3.90
YPCB048	8.1	Yellow Perch	*Perca flavescens*	-28.36	6.65	43.61	11.72	3.72
YPCB048	8.1	Yellow Perch	*Perca flavescens*	-28.13	6.95	45.73	13.09	3.49
YPCB049	8.3	Yellow Perch	*Perca flavescens*	-27.00	7.20	45.10	11.82	3.82
YPCB050	8.4	Yellow Perch	*Perca flavescens*	-27.95	6.95	43.55	11.41	3.82
YPCB051	7.7	Yellow Perch	*Perca flavescens*	-28.95	6.96	46.21	12.34	3.74
YPCB052	6.3	Yellow Perch	*Perca flavescens*	-28.42	6.61	45.74	11.66	3.92
YPCB053	8.5	Yellow Perch	*Perca flavescens*	-26.82	7.41	44.44	12.68	3.51
YPGR001	7.9	Yellow Perch	*Perca flavescens*	-32.84	7.56	40.45	9.68	4.18
YPGR002	4.3	Yellow Perch	*Perca flavescens*	-32.63	7.21	44.95	12.59	3.57
YPLO019	6.7	Yellow Perch	*Perca flavescens*	-30.81	6.94	46.45	14.06	3.30

Specimen Code	TL (cm)	Common name	Scientific name	d13C	d15N	%C	%N	C/N
YPLO020	7.8	Yellow Perch	*Perca flavescens*	-28.30	7.41	46.39	14.17	3.27
YPLO021	11.3	Yellow Perch	*Perca flavescens*	-32.53	6.79	45.90	14.36	3.20
YPLO022	7.3	Yellow Perch	*Perca flavescens*	-30.81	7.04	44.52	13.25	3.36
YPLO023	7.2	Yellow Perch	*Perca flavescens*	-31.74	6.95	46.38	14.04	3.30
YPLO024	5.7	Yellow Perch	*Perca flavescens*	-33.09	7.72	46.82	13.75	3.41
YPLO025	6.7	Yellow Perch	*Perca flavescens*	-30.60	5.86	43.53	11.20	3.89
YPLO026	6.5	Yellow Perch	*Perca flavescens*	-30.71	7.24	46.59	12.46	3.74
YPLO027	5.6	Yellow Perch	*Perca flavescens*	-30.96	6.26	44.62	12.55	3.55
YPLO028	7.4	Yellow Perch	*Perca flavescens*	-29.98	6.27	44.76	11.62	3.85
YPLO028	7.4	Yellow Perch	*Perca flavescens*	-30.06	6.10	46.70	11.82	3.95
YPLO029	6.4	Yellow Perch	*Perca flavescens*	-31.48	6.60	47.43	11.72	4.05
YPLO030	5.8	Yellow Perch	*Perca flavescens*	-30.53	5.95	47.09	12.75	3.69
YPLO031	6.6	Yellow Perch	*Perca flavescens*	-30.99	6.39	44.58	11.11	4.01
StyscudBD001		Zebra Clubtail	*Stylurus scudderi*	-30.13	4.75	44.85	11.47	3.91
ZOBD052		Zooplankton	n/a	-34.24	2.47	43.97	9.53	4.61
ZOBD052		Zooplankton	n/a	-34.18	2.53	44.04	9.36	4.70
ZOBD053		Zooplankton	n/a	-34.11	3.07	43.62	9.37	4.66
ZOBD054		Zooplankton	n/a	-33.54	2.76	27.20	5.79	4.70
ZOGR062		Zooplankton	n/a	-35.04	3.31	43.34	9.06	4.79
ZOLO010		Zooplankton	n/a	-34.52	2.51	44.06	9.23	4.77
ZOLO011		Zooplankton	n/a	-34.32	2.33	21.69	4.50	4.81

Specimen Code	TL (cm)	Common name	Scientific name	d13C	d15N	%C	%N	C/N
ZOCB012		Zooplankton	n/a	-33.99	2.27	39.63	8.26	4.80
ZOCB016		Zooplankton	n/a	-32.83	2.16	38.60	7.58	5.09
Elm2020Oct07KNP-001			n/a	-31.17	3.28	56.90	8.96	6.35
Elm2020Oct07KNP-002			n/a	-31.74	3.18	55.82	9.07	6.16
Meg2020Oct07KNP-001			n/a	-29.92	4.59	53.62	9.22	5.82
Meg2020Oct07KNP-002			n/a	-29.02	4.21	53.83	9.55	5.63
Meg2020Oct07KNP-003			n/a	-29.56	4.29	51.40	10.14	5.07
Meg2020Oct07KNP-003			n/a	-29.58	4.33	50.23	10.25	4.90
Meg2020Oct07KNP-004			n/a	-28.94	4.18	50.06	10.55	4.75
Odo2020Oct07KNP-001			n/a	-27.02	3.21	45.97	11.83	3.89
Ple2020Oct07KNP-001			n/a	-30.71	4.29	54.82	9.82	5.58
TroHydrCB001			n/a	-30.78	5.22	58.99	7.48	7.89

Appendix B: Zooplankton Data

IdentaZoop

ZOOPLANKTON BENCH SHEET

Sample ID	ZOL0009	**Comments:** Entire sample was processed. No size class 4 found.	**ZEBRA count ID**	SC04993	
Station #	LO1		**Taxonomist**	Lynne M. Witty	
Lake code	LOO		**Count date**	Monday, November 8, 2021	
Lake name	Loon Lake		**Working volume (mL)**	250	
Sampling date	June 10, 2021				
Sampling time			**Size class**	**Count category**	**Total count**
Gear	12" tow net		1	< 0.6 mm (nauplii)	0
Mesh size (µm)	243		2	< 0.6 mm	150
Haul length (m)	7.2		3	≥ 0.6 mm to < 1.2 mm	153
Sample volume (L)	2626.77		4	≥ 1.2mm	0
			Total count in SSV		**303**

***Mean weight data is derived from the associated ZEBRA count, except for Zebra mussel veligers (ZEBRA code 650) for which weights are calculated using the new DFO equation (2018).**

ZEBRA species code	Size class	Taxa	Total count in SSV	Subsample volume (SSV) (mL)	Fraction analyzed	Total # in entire sample	Density (#/m³)	Mean length (mm)	Mean weight (µg)	ZEBRA Biomass (mg/m³)
135	2	Holopedium gibberum	103	15	0.060	1717	653.528	0.410	0.894	0.584
135	3	Holopedium gibberum	103	65	0.260	396	150.814	0.666	3.303	0.498
150	2	Eubosmina (Eubosmina) longispina	29	15	0.060	483	184.003	0.290	1.175	0.216
152	3	Diaphanosoma birgei	3	65	0.260	12	4.393	0.750	3.749	0.016
188	2	Bosmina (Bosmina) longirostris	17	15	0.060	283	107.864	0.287	1.139	0.123
201	2	Calanoid copepodid	1	15	0.060	17	6.345	0.393	0.554	0.004
201	3	Calanoid copepodid	2	65	0.260	8	2.928	0.666	2.021	0.006
204	3	Leptodiaptomus minutus	29	65	0.260	112	42.462	0.773	2.944	0.125
211	3	Epischura sp. copepodid	3	65	0.260	12	4.393	0.926	4.857	0.021
301	3	Cyclopoid copepodid	3	65	0.260	12	4.393	0.762	2.889	0.013
309	3	Mesocyclops edax	9	65	0.260	35	13.178	0.806	3.383	0.045
719	3	Epischura nordenskioldi	1	65	0.260	4	1.464	1.172	8.125	0.012
TOTALS			**303**			**3088**	**1175.764**		**35.035**	**1.663**

IDENTAZOOP

ZOOPLANKTON BENCH SHEET

Sample ID	ZOLO013	**Comments:** Entire sample was processed.	**ZEBRA count ID**	SC04994	
Station #	LO1		**Taxonomist**	Lynne M. Witty	
Lake code	LOO		**Count date**	Monday, November 8, 2021	
Lake name	Loon Lake		**Working volume (mL)**	250	
Sampling date	June 10, 2021				
Sampling time			**Size class**	**Count category**	**Total count**
Gear	12" tow net		1	< 0.6 mm (nauplii)	24
Mesh size (µm)	53		2	< 0.6 mm	130
Haul length (m)	7.2		3	≥ 0.6 mm to < 1.2 mm	34
Sample volume (L)	2626.77		4	≥ 1.2mm	0
			Total count in SSV		**188**

***Mean weight data is derived from the associated ZEBRA count, except for Zebra mussel veligers (ZEBRA code 650) for which weights are calculated using the new DFO equation (2018).**

ZEBRA species code	Size class	Taxa	Total count in SSV	Subsample volume (SSV) (mL)	Fraction analyzed	Total # in entire sample	Density (#/m³)	Mean length (mm)	Mean weight (µg)	ZEBRA Biomass (mg/m³)
133	2	Eubosmina (Neobosmina) tubicen	16	30	0.120	133	50.759	0.258	0.875	0.044
135	2	Holopedium gibberum	1	30	0.120	8	3.172	0.349	0.457	0.001
150	2	Eubosmina (Eubosmina) longispina	29	30	0.120	242	92.001	0.236	0.722	0.066
152	2	Diaphanosoma birgei	1	30	0.120	8	3.172	0.327	1.570	0.005
188	2	Bosmina (Bosmina) longirostris	45	30	0.120	375	142.761	0.249	0.815	0.116
201	2	Calanoid copepodid	6	30	0.120	50	19.035	0.486	0.957	0.018
201	3	Calanoid copepodid	22	250	1.000	22	8.375	0.658	1.988	0.017
204	3	Leptodiaptomus minutus	7	250	1.000	7	2.665	0.737	2.599	0.007
211	2	Epischura sp. copepodid	27	30	0.120	225	85.657	0.361	0.469	0.040
215	1	Calanoid nauplius	20	30	0.120	167	63.449	0.223	0.235	0.015
301	2	Cyclopoid copepodid	5	30	0.120	42	15.862	0.366	0.482	0.008
301	3	Cyclopoid copepodid	1	250	1.000	1	0.381	0.676	2.099	0.001
309	3	Mesocyclops edax	4	250	1.000	4	1.523	0.689	2.219	0.003
313	1	Cyclopoid nauplius	4	30	0.120	33	12.690	0.221	0.220	0.003
TOTALS			**188**			**1317**	**501.503**		**15.707**	**0.345**

IDENTAZOOP

ZOOPLANKTON BENCH SHEET

Sample ID	ZOGR061	**ZEBRA count ID**	SC04991
Station #	GR1	**Taxonomist**	Lynne M. Witty
Lake code	GFN	**Count date**	Friday, November 5, 2021
Lake name	Grafton Lake	**Working volume (mL)**	250
Sampling date	June 25, 2021		
Sampling time			
Gear	12" tow net		
Mesh size (µm)	243		
Haul length (m)	4.1		
Sample volume (L)	1495.80		

Comments: Entire sample was processed.

Size class	Count category	Total count
1	<0.6 mm (nauplii)	5
2	<0.6 mm	145
3	≥0.6 mm to < 1.2 mm	164
4	≥1.2mm	40
Total count in SSV		**354**

*Mean weight data is derived from the associated ZEBRA count, except for Zebra mussel veligers (ZEBRA code 650) for which weights are calculated using the new DFO equation (2018).

ZEBRA species code	Size class	Taxa	Total count in SSV	Subsample volume (SSV) (mL)	Fraction analyzed	Total # in entire sample	Density (#/m³)	Mean length (mm)	Mean weight (µg)	ZEBRA Biomass (mg/m³)
120	2	Daphnia (Daphnia) catawba	82	10	0.040	2050	1370.504	0.530	0.834	1.143
120	3	Daphnia (Daphnia) catawba	142	10	0.040	3550	2373.312	0.787	2.721	6.459
120	4	Daphnia (Daphnia) catawba	24	250	1.000	24	16.045	1.272	9.926	0.159
135	2	Holopedium gibberum	2	10	0.040	50	33.427	0.343	0.434	0.015
135	4	Holopedium gibberum	6	250	1.000	6	4.011	1.290	24.597	0.099
138	4	Leptodora kindtii	4	250	1.000	4	2.674	2.748	5.575	0.015
145	4	Sida crystallina	2	250	1.000	2	1.337	1.336	11.500	0.015
152	2	Diaphanosoma birgei	10	10	0.040	250	167.135	0.476	2.329	0.389
152	3	Diaphanosoma birgei	3	10	0.040	75	50.140	0.700	3.486	0.175
201	2	Calanoid copepodid	30	10	0.040	750	501.404	0.482	0.948	0.475
201	3	Calanoid copepodid	12	10	0.040	300	200.562	0.684	2.197	0.441
204	3	Leptodiaptomus minutus	6	10	0.040	150	100.281	0.726	2.536	0.254
211	2	Epischura sp. copepodid	20	10	0.040	500	334.269	0.362	0.481	0.161
215	1	Calanoid nauplius	5	10	0.040	125	83.567	0.221	0.229	0.019
301	2	Cyclopoid copepodid	1	10	0.040	25	16.713	0.336	0.377	0.006
301	3	Cyclopoid copepodid	1	10	0.040	25	16.713	0.764	2.835	0.047
719	4	Epischura nordenskioldi	4	250	1.000	4	2.674	1.386	12.572	0.034
TOTALS			**354**			**7890**	**5274.769**		**83.578**	**9.906**

IDENTAZOOP

ZOOPLANKTON BENCH SHEET

Sample ID	ZOGR060	**Comments:** Entire sample was processed.	**ZEBRA count ID**	SC04992	
Station #	GR1		**Taxonomist**	Lynne M. Witty	
Lake code	GFN		**Count date**	Friday, November 5, 2021	
Lake name	Grafton Lake		**Working volume (mL)**	250	
Sampling date	June 25, 2021				
Sampling time			**Size class**	**Count category**	**Total count**
Gear	12" tow net		1	< 0.6 mm (nauplii)	30
Mesh size (µm)	53		2	< 0.6 mm	143
Haul length (m)	4.1		3	≥ 0.6 mm to < 1.2 mm	24
Sample volume (L)	1495.80		4	≥ 1.2mm	0
			Total count in SSV		**197**

***Mean weight data is derived from the associated ZEBRA count, except for Zebra mussel veligers (ZEBRA code 650) for which weights are calculated using the new DFO equation (2018).**

ZEBRA species code	Size class	Taxa	Total count in SSV	Subsample volume (SSV) (mL)	Fraction analyzed	Total # in entire sample	Density (#/m³)	Mean length (mm)	Mean weight (µg)	ZEBRA Biomass (mg/m³)
120	3	Daphnia (Daphnia) catawba	2	250	1.000	2	1.337	0.682	1.703	0.002
201	2	Calanoid copepodid	74	10	0.040	1850	1236.796	0.409	0.634	0.784
201	3	Calanoid copepodid	17	250	1.000	17	11.365	0.627	1.746	0.020
204	3	Leptodiaptomus minutus	5	250	1.000	5	3.343	0.679	2.128	0.007
211	2	Epischura sp. copepodid	64	10	0.040	1600	1069.662	0.341	0.398	0.426
215	1	Calanoid nauplius	24	5	0.020	1200	802.246	0.211	0.212	0.170
301	2	Cyclopoid copepodid	4	10	0.040	100	66.854	0.367	0.476	0.032
313	1	Cyclopoid nauplius	6	5	0.020	300	200.562	0.186	0.165	0.033
338	2	Tropocyclops extensus	1	10	0.040	25	16.713	0.494	0.971	0.016
TOTALS			**197**			**5099**	**3408.878**		**8.433**	**1.491**

IDENTAZOOP

ZOOPLANKTON BENCH SHEET

Sample ID	ZOBD051	**Comments:**	**ZEBRA count ID**	SC04989	
Station #	CB1		**Taxonomist**	Lynne M. Witty	
Lake code	BDW		**Count date**	Thursday, November 4, 2021	
Lake name	Big Dam West Lake		**Working volume (mL)**	250	
Sampling date	June 18, 2021				
Sampling time			**Size class**	**Count category**	**Total count**
Gear	12" tow net		1	<0.6 mm (nauplii)	2
Mesh size (µm)	243		2	<0.6 mm	155
Haul length (m)	5.2		3	≥0.6 mm to <1.2 mm	172
Sample volume (L)	1897.11		4	≥1.2mm	158
			Total count in SSV		**487**

*Mean weight data is derived from the associated ZEBRA count, except for Zebra mussel veligers (ZEBRA code 650) for which weights are calculated using the new DFO equation (2018).

ZEBRA species code	Size class	Taxa	Total count in SSV	Subsample volume (SSV) (mL)	Fraction analyzed	Total # in entire sample	Density (#/m³)	Mean length (mm)	Mean weight (µg)	ZEBRA Biomass (mg/m³)
120	2	Daphnia (Daphnia) catawba	7	10	0.040	175	92.246	0.518	0.786	0.072
120	3	Daphnia (Daphnia) catawba	23	12	0.048	479	252.577	0.767	2.446	0.618
135	2	Holopedium gibberum	114	10	0.040	2850	1502.285	0.521	1.622	2.437
135	3	Holopedium gibberum	26	12	0.048	542	285.522	0.636	2.848	0.813
138	4	Leptodora kindtii	3	112	0.448	7	3.530	4.257	20.896	0.074
150	2	Eubosmina (Eubosmina) longispina	9	10	0.040	225	118.601	0.302	1.333	0.158
152	2	Diaphanosoma birgei	3	10	0.040	75	39.534	0.559	2.751	0.109
152	3	Diaphanosoma birgei	8	12	0.048	167	87.853	0.707	3.526	0.310
188	2	Bosmina (Bosmina) longirostris	7	10	0.040	175	92.246	0.254	0.849	0.078
201	2	Calanoid copepodid	1	10	0.040	25	13.178	0.429	0.686	0.009
201	3	Calanoid copepodid	9	12	0.048	188	98.835	0.709	2.412	0.238
204	3	Leptodiaptomus minutus	17	12	0.048	354	186.687	0.802	3.303	0.617
211	2	Epischura sp. copepodid	6	10	0.040	150	79.068	0.482	0.916	0.072
211	3	Epischura sp. copepodid	12	12	0.048	250	131.779	0.946	5.125	0.675
211	4	Epischura sp. copepodid	5	112	0.448	11	5.883	1.274	10.021	0.059
215	1	Calanoid nauplius	1	10	0.040	25	13.178	0.251	0.284	0.004
301	2	Cyclopoid copepodid	6	10	0.040	150	79.068	0.447	0.817	0.065
301	3	Cyclopoid copepodid	6	12	0.048	125	65.890	0.670	2.075	0.137
309	2	Mesocyclops edax	2	10	0.040	50	26.356	0.554	1.297	0.034
309	3	Mesocyclops edax	5	12	0.048	104	54.908	0.884	4.487	0.246
309	4	Mesocyclops edax	4	112	0.448	9	4.706	1.239	9.320	0.044
313	1	Cyclopoid nauplius	1	10	0.040	25	13.178	0.211	0.203	0.003
719	3	Epischura nordenskioldi	3	12	0.048	63	32.945	1.175	8.171	0.269
719	4	Epischura nordenskioldi	146	112	0.448	326	171.784	1.323	11.006	1.891
733	3	Skistodiaptomus pygmaeus	63	12	0.048	1313	691.842	0.935	4.716	3.263
TOTALS			**487**			**7861**	**4143.677**		**101.897**	**12.295**

IDENTAZOOP

ZOOPLANKTON BENCH SHEET

Sample ID	ZOBD050	**Comments:** Entire sample was processed.	**ZEBRA count ID**	SC04990
Station #	CB1		**Taxonomist**	Lynne M. Witty
Lake code	BDW		**Count date**	Friday, November 5, 2021
Lake name	Big Dam West Lake		**Working volume (mL)**	100
Sampling date	June 18, 2021			
Sampling time				
Gear	12" tow net			
Mesh size (µm)	53			
Haul length (m)	5.2			
Sample volume (L)	1897.11			

Size class	Count category	Total count
1	< 0.6 mm (nauplii)	22
2	< 0.6 mm	128
3	≥ 0.6 mm to < 1.2 mm	25
4	≥ 1.2mm	1
Total count in SSV		**176**

*Mean weight data is derived from the associated ZEBRA count, except for Zebra mussel veligers (ZEBRA code 650) for which weights are calculated using the new DFO equation (2018).

ZEBRA species code	Size class	Taxa	Total count in SSV	Subsample volume (SSV) (mL)	Fraction analyzed	Total # in entire sample	Density (#/m³)	Mean length (mm)	Mean weight (µg)	ZEBRA Biomass (mg/m³)
135	2	Holopedium gibberum	9	25	0.250	36	18.976	0.327	0.501	0.010
152	2	Diaphanosoma birgei	3	25	0.250	12	6.325	0.373	1.803	0.011
188	2	Bosmina (Bosmina) longirostris	11	25	0.250	44	23.193	0.218	0.610	0.014
201	2	Calanoid copepodid	32	25	0.250	128	67.471	0.456	0.815	0.055
201	3	Calanoid copepodid	12	100	1.000	12	6.325	0.668	2.065	0.013
204	3	Leptodiaptomus minutus	4	100	1.000	4	2.108	0.677	2.121	0.004
211	2	Epischura sp. copepodid	24	25	0.250	96	50.603	0.359	0.452	0.023
211	3	Epischura sp. copepodid	1	100	1.000	1	0.527	1.047	6.156	0.003
215	1	Calanoid nauplius	16	25	0.250	64	33.736	0.170	0.153	0.005
301	2	Cyclopoid copepodid	46	25	0.250	184	96.990	0.411	0.667	0.065
301	3	Cyclopoid copepodid	5	100	1.000	5	2.636	0.666	2.078	0.005
313	1	Cyclopoid nauplius	6	25	0.250	24	12.651	0.204	0.194	0.002
338	2	Tropocyclops extensus	3	25	0.250	12	6.325	0.442	0.751	0.005
719	4	Epischura nordenskioldi	1	100	1.000	1	0.527	1.218	8.934	0.005
733	3	Skistodiaptomus pygmaeus	3	100	1.000	3	1.581	0.825	3.433	0.005
TOTALS			**176**			**626**	**329.976**		**30.733**	**0.226**

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ZOOPLANKTON BENCH SHEET

Sample ID	ZOCB002	**ZEBRA count ID**	SC04987
Station #	CB1	**Taxonomist**	Lynne M. Witty
Lake code	COB	**Count date**	Thursday, November 4, 2021
Lake name	Cobrielle Lake	**Working volume (mL)**	250
Sampling date	June 4, 2021		
Sampling time			
Gear	12" tow net		
Mesh size (µm)	243		
Haul length (m)	5.2		
Sample volume (L)	1897.11		

Comments: Entire sample was processed.

Size class	Count category	Total count
1	<0.6 mm (nauplii)	11
2	<0.6 mm	183
3	≥0.6 mm to <1.2 mm	99
4	≥1.2mm	0
Total count in SSV		**293**

*Mean weight data is derived from the associated ZEBRA count, except for Zebra mussel veligers (ZEBRA code 650) for which weights are calculated using the new DFO equation (2018).

ZEBRA species code	Size class	Taxa	Total count in SSV	Subsample volume (SSV) (mL)	Fraction analyzed	Total # in entire sample	Density (#/m³)	Mean length (mm)	Mean weight (µg)	ZEBRA Biomass (mg/m³)
120	2	Daphnia (Daphnia) catawba	17	90	0.360	47	24.892	0.526	0.821	0.020
120	3	Daphnia (Daphnia) catawba	62	250	1.000	62	32.681	0.774	2.497	0.082
135	3	Holopedium gibberum	1	250	1.000	1	0.527	0.772	5.099	0.003
150	2	Eubosmina (Eubosmina) longispina	146	90	0.360	406	213.775	0.282	1.133	0.242
152	2	Diaphanosoma birgei	2	90	0.360	6	2.928	0.486	2.380	0.007
152	3	Diaphanosoma birgei	2	250	1.000	2	1.054	0.705	3.513	0.004
188	2	Bosmina (Bosmina) longirostris	5	90	0.360	14	7.321	0.313	1.338	0.010
201	2	Calanoid copepodid	4	90	0.360	11	5.857	0.514	1.150	0.007
201	3	Calanoid copepodid	7	250	1.000	7	3.690	0.655	1.956	0.007
204	2	Leptodiaptomus minutus	1	90	0.360	3	1.464	0.593	1.522	0.002
204	3	Leptodiaptomus minutus	24	250	1.000	24	12.651	0.699	2.303	0.029
211	2	Epischura sp. copepodid	8	90	0.360	22	11.714	0.425	0.681	0.008
211	3	Epischura sp. copepodid	3	250	1.000	3	1.581	0.668	2.039	0.003
215	1	Calanoid nauplius	7	90	0.360	19	10.250	0.189	0.180	0.002
313	1	Cyclopoid nauplius	4	90	0.360	11	5.857	0.200	0.191	0.001
TOTALS			**293**			**638**	**336.242**		**26.802**	**0.427**

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ZOOPLANKTON BENCH SHEET

Sample ID	ZOCB005	**Comments:** Entire sample was processed.	**ZEBRA count ID**	SC04988	
Station #	CB1		**Taxonomist**	Lynne M. Witty	
Lake code	COB		**Count date**	Thursday, November 4, 2021	
Lake name	Cobrielle Lake		**Working volume (mL)**	250	
Sampling date	June 4, 2021				
Sampling time			**Size class**	**Count category**	**Total count**
Gear	12" tow net		1	<0.6 mm (nauplii)	50
Mesh size (µm)	53		2	<0.6 mm	152
Haul length (m)	5.2		3	≥0.6 mm to <1.2 mm	4
Sample volume (L)	1897.11		4	≥1.2mm	0
			Total count in SSV		**206**

*Mean weight data is derived from the associated ZEBRA count, except for Zebra mussel veligers (ZEBRA code 650) for which weights are calculated using the new DFO equation (2018).

ZEBRA species code	Size class	Taxa	Total count in SSV	Subsample volume (SSV) (mL)	Fraction analyzed	Total # in entire sample	Density (#/m³)	Mean length (mm)	Mean weight (µg)	ZEBRA Biomass (mg/m³)
109	2	Alona sp.	1	250	1.000	1	0.527	0.356	1.822	0.001
118	2	Chydorus sphaericus	1	250	1.000	1	0.527	0.214	0.667	0.000
119	2	Daphnia (Daphnia) ambigua	1	250	1.000	1	0.527	0.565	0.989	0.001
150	2	Eubosmina (Eubosmina) longispina	84	250	1.000	84	44.278	0.227	0.670	0.030
188	2	Bosmina (Bosmina) longirostris	3	250	1.000	3	1.581	0.296	1.203	0.002
201	2	Calanoid copepodid	30	250	1.000	30	15.814	0.392	0.606	0.010
201	3	Calanoid copepodid	3	250	1.000	3	1.581	0.634	1.800	0.003
209	3	Leptodiaptomus siciloides	1	250	1.000	1	0.527	1.072	6.521	0.003
211	2	Epischura sp. copepodid	27	250	1.000	27	14.232	0.341	0.405	0.006
215	1	Calanoid nauplius	29	80	0.320	91	47.770	0.212	0.216	0.010
301	2	Cyclopoid copepodid	4	250	1.000	4	2.108	0.386	0.558	0.001
313	1	Cyclopoid nauplius	21	80	0.320	66	34.592	0.199	0.187	0.006
338	2	Tropocyclops extensus	1	250	1.000	1	0.527	0.461	0.820	0.000
TOTALS			**206**			**312**	**164.592**		**16.464**	**0.073**