Great Lake Fisheries Trust Final Report (September 2018)

Grant No. 2015.1550

Project Abstract

The New Lake Michigan Food Web:

Establishing links between nearshore food sources and pelagic piscivores

Purpose:

The primary objective of this project was to better characterize the diets of large-bodied piscivores in Lake Michigan following major ecosystem changes in the past 10-15 years. More specifically, broad scale changes in nutrient-algal dynamics, energy flows, and relative species abundance have been observed including establishment of invasive dreissenid mussels and round goby. This project aimed to address several questions related to the trophic ecology and management of Lake Michigan piscivores following these changes. Questions were: 1) Does the ability to utilize nearshore energy sources vary among species? 2) Within a given species, does the dependence on nearshore energy sources vary with location? and 3) Do spatial and temporal differences in dependence on nearshore energy lead to differences in fish condition? To achieve these objectives, 5 major tasks were completed: 1) collection of fish for regional comparisons of upper food web structure, 2) regional comparison of upper food web structure, 3) confirmation of spatial trends in nearshore trophic structure, 4) determine long-term trends in upper food web structure, and 5) analysis, presentation and publication of data. This work has ultimately improved understanding of energy pathways and trophic linkages in Lake Michigan and will be used to inform inputs to statistical catch at age models and ecosystem models currently used for management.

Material and Methods:

Stomach Content Analysis

Once stomachs were thawed, stomach contents were removed for processing. All fish prey were identified to species, except for sculpins, which were identified to family. Each individual fish prey were measured to the nearest 1 mm standard or vertebral length depending on digestion. Highly digested fish prey were identified using vertebrae (Elliot et al., 1996) and cleithra (Traynor et al., 2010). Depending on digestion, total lengths were estimating from standard, vertebral, or cleithra (if attached to partial vertebrae) based on published conversion formulae (Dub & Czesny, 2016; Elliot et al., 1996; Knight et al., 1984; Kornis et al., 2012; J. Jonas, MDNR, pers. comm.). Invertebrate prey were identified to the lowest taxonomic level. Each prey group were wet and dry weighed to the nearest 0.01 g. We examined the diet compositions of Lake Michigan piscivores by estimating the mean percent diet composition by weight and the average weight of prey categories found in stomachs (Elliot et al., 1996).

Stable Isotope Analysis

Stable isotope measurements were made using the methods described in Turschak and Bootsma (2015), and Ngochera and Bootsma (2011). Briefly, isotope concentrations were measured using an isotope ratio mass spectrometer (Finnigan MAT delta S SIR-MS) with elemental analyzer front end and ConFlo II interface. Carbon calibration was done with NIST standard RM 8542 (sucrose, δ^{13}C=−10.47) and a NIST-traceable standard (glycine, δ^{13}C=−33.63). Nitrogen calibration was with NIST standard RM 8547 (IAEA-N1 ammonium sulfate, δ^{1}5N=0.4), NIST standard RM 8548 (IAEA-N_2 ammonium sulfate, δ^{15}N=20.3), and a NIST-traceable ammonium chloride standard (δ^{15}N=−8.9). During sample runs, an acetanilide control sample was run every twelfth sample and analyzed for ^{13}C:^{12}C and ^{15}N:^{14}N ratios. Instrument precision was ±0.2‰ for both C and N isotopes based upon acetanilide controls. All stable isotope results are expressed in δ notation (i.e. δ^{13}C and δ^{15}N) as per mil (‰) differences between the isotope ratio of the sample and that of the international standard (PDB carbonate and atmospheric air for δ^{13}C and δ^{15}N, respectively). Taxa specific lipid-corrections were applied to δ^{13}C values following the equations described in Turschak and Bootsma (2015) to reduce variability associated with consumer tissue lipid content.

We quantified the isotopic niche overlap and diets of large bodied piscivores across sites using a Bayesian statistical approaches. The “nicheROVER” package (Swanson et al. 2015) built for R was used to perform pairwise comparisons of isotopic niche overlap among species. Likewise, the “MixSIAR” package (Stock and Semmens 2013) built for R was used to quantify dietary proportions of prey sources using δ^{13}C and δ^{15}N as tracers, region as a random effect factor, and total length as a covariate. Potential prey sources for each species were determined as the 4-5 most abundant prey sources from stomach content analyses. Isotope ratios of round goby prey were site-specific while a lake-wide average was used for other species due to lower regional variation (Turschak et al. 2018). In addition to source isotope data, trophic enrichment factors (mean ± sd) of 0.4 ± 1.3 for δ^{13}C and 3.4 ± 1.0 for δ^{15}N were provided as model inputs (Post 2002). We used informed prior probability distributions based on observed stomach content data with three Markov chain Monte Carlo (MCMC) simulations of 1,000,000 iterations. The burn-in period for each iteration chain was set at 500,000 and the subsequent values were thinned by a factor of 500. Chain convergence was checked using the Gelman-Rubin and Geweke diagnostic tests. If chains had not converged, a second model run was performed with three chains of length 3,000,000, burn-in period of 1,500,000, and thinned by a factor of 500.

Fatty Acid Analysis

For fatty acid analysis, belly flap samples or whole prey fish were homogenized separately. Lipids were extracted from each homogenate according to Folch et al. (1957) allowing a measure of the lipid content of each tissue sample or whole prey fish. Fatty acid methyl esters (FAME) were prepared according to Metcalfe and Schmitz (1961), separated by gas chromatography/mass spectrometry (Agilent 7890A GC and 5975C inert XL EI/CI MSD, Agilent Technologies, Inc., Santa Clara, California, USA) and quantified as previously described (Czesny et al. 2011). Fatty acid data are expressed as percent composition by mass for each individual (Happel et al. 2017a).

A ratio of 18:1n-9 to 16:1n-7 was used as a relative indicator of alewife vs. round goby consumption, visualized as mean and 95% confidence intervals by region. Whole profiles of fatty acids were analyzed using multivariate techniques in R. nMDS coordinates were calculated for all samples, predators and prey species, and extracted. This technique allows one to summarize how similar multivariate data points to each other in a 2-dimensional plane and is a useful exploratory technique. From this we summarize prey data by regions within the lake, illustrating that within prey species variability is smaller than between the prey species. The difference between species was statistically tested using linear discriminant function analysis (LDA).

We then summarized alewife, bloater, rainbow smelt, and round goby as lake-wide averages and used these as reference points to visually explore ontogenetic shifts in salmon fatty acid profiles. We tested for seasonal and regional differences using linear discriminant analysis for each salmon species separately. For brevity we include here only the analysis of Chinook Salmon and Lake Trout, arguably the species of most interest.

Stomach Content Analysis Results (Table 1a)

Brown Trout

Alewife and round goby were clearly the dominant prey items for brown trout, by both percent diet composition (calculated as the mean of individual proportional diet composition; alewife: 38%; round goby: 21%) and by mean weight per stomach (alewife: 2.1 g; round goby: 3.1 g; Fig. 1). Round goby were primarily consumed in the spring on the eastern side of Lake Michigan (Fig. 2).

Chinook Salmon

Chinook salmon feed almost exclusively on alewife (proportional diet composition: 73%; mean g/stomach: 9.0 g; Fig. 1). Chinook salmon did consume other prey items, including *Bythotrephes*, *Mysis,* yellow perch, and bloater, but these made minimal contributions.

Coho Salmon

Coho salmon primarily consumed alewife (54%), but aquatic invertebrates (Mysis: 13%; Bythotrephes: 17%; Fig. 1) contributed considerable proportions to the Coho salmon diet compositions. Despite aquatic invertebrates contributing a large proportion to the mean diet composition of Coho salmon, the mean weight of all invertebrates (<1 g) found in Coho salmon stomachs was minimal compared to alewife (10.8 g; Fig. 1). Mysis were primarily consumed by small Coho salmon (<600 mm) in the southeast region in the spring, whereas Bythotrephes were consumed mainly by large Coho salmon (>600 mm) in the late summer/fall (Fig. 3).

Lake Trout

Lake trout had feeding patterns similar to those of brown trout, with alewife (56% and 9.3 g/stomach) and round goby (30% and 3.2 g/stomach; Fig. 1) being the dominant prey items for lake trout. Also, similar to brown trout, the majority of round goby consumption occurred in the spring on the eastern half of the lake (Fig. 2).

Steelhead

Steelhead consumed primarily alewife (37%) and terrestrial insects (33%; Fig. 1). Although alewife and terrestrial insects contributed similar proportions to the overall diet composition, the mean weight of alewife in steelhead stomachs (8.7 g) was much greater than terrestrial insects (1.6 g; Fig. 2).

Across-Salmonine Diet Patterns

Although the different salmonine species had distinct feeding behaviors, there were consistent spatio-temporal feeding patterns across the five salmonine species. In the spring, alewife were the dominant prey item for all salmonines on the western half of Lake Michigan (Fig. 2). In the spring on the eastern side, salmonines consumed primarily round goby (brown trout and lake trout), aquatic invertebrates (Coho salmon), and terrestrial invertebrates (steelhead) instead of alewife (Fig. 2). These feeding patterns did not occur in the late summer/fall (Fig. 2). *Mysis* and yellow perch were primarily consumed in the southern regions in the spring and fall, respectively. Bloater and *Bythotrephes* were consumed most in the late summer/fall (Fig. 2). Generally, aquatic and terrestrial invertebrates were consumed more by small salmonines (<600 mm; Fig. 3).

Alewife Lengths Consumed by Salmonines

The length distribution of alewife consumed by salmonines showed a bimodal pattern with the majority of alewife consumed being less than 120 mm (Fig. 4). Length frequencies of consumed alewife were similar to length frequencies of alewife collected in USGS fall bottom trawls (Fig. 4). Small alewives (<120 mm) were most commonly found in stomachs collected in western regions, whereas large alewife (>120 mm) were most common in eastern regions (Fig. 5). On average, steelhead consumed the smallest alewife (95.3 mm), whereas the other four species consumed larger alewife.

Burbot

The primary prey item for burbot was round goby (2016: 54%; 2017: 43%), but alewife, aquatic invertebrates (i.e., crayfish, dreissenid mussels, and *Mysis*), sculpin, and other fish (i.e., fish eggs, rainbow smelt, white sucker, and yellow perch) contributed to their diet composition (Fig. 6). Although round goby was the dominant prey item across Lake Michigan, there was some spatial variation in the consumption patterns of other prey items, such as the consumption of sculpins in the southern regions (only in 2016) and alewife in the eastern regions (Fig. 6).

Lake Whitefish

Lake whitefish primarily consumed aquatic invertebrates, although the types of aquatic invertebrates consumed varied by region of capture. In the northeast, lake whitefish primarily consumed chironomids (larvae: 12%; pupae: 56%), but also consumed round goby (9%; Fig. 7). In the southeast, lake whitefish stomachs consisted of mostly gastropods (19%) and *Mysis* (50%; Fig. 7). Although we collected several lake whitefish from the northwest region of Lake Michigan, 25 out of the 27 stomachs analyzed were empty (Fig. 7), so we do not have an accurate description of their foraging patterns in that region of the lake.

Stable Isotope Analysis Results (Table 1b)

Trophic Position and Isotopic Niche Overlap

Stable C and N isotope ratios of most Lake Michigan salmonines were relatively similar (Fig. 8). Lake wide mean δ^{13}C was between -23.5‰ and -22.5‰ for all salmonines (Fig. 8 and Fig. 9). Mean δ^{15}N was between 10‰ and 11.5‰ for all salmonines except lake trout which had a greater mean δ^{15}N (13‰; Fig. 8 and Fig. 9). Because of their higher δ^{15}N, lake trout exhibited the lowest probability of niche overlap with other salmonines. Lake trout and Brown Trout had broader ranges of δ^{13}C which resulted in a relatively large isotopic niche area as revealed by 95% Bayesian ellipse area. Steelhead also exhibited a relatively large niche area although more of this variation fell along the δ^{15}N axis. Chinook salmon had an intermediate niche area and a high probability of overlap with other salmonines. Although Coho salmon had the smallest niche area, the probability of niche overlap with other salmonines was lower because of low δ^{15}N in this species.

Diet Portions

Regional differences in diet patterns were smaller than differences associated with ontogeny for all salmonines except lake trout (Figs. 10-14). As with diet analyses, alewives were the dominant prey for all species. Chinook salmon appeared to transition from a diet dominated by small alewives to one comprised of larger alewives and bloater as total length increased (Fig 10). In contrast, Coho salmon and Steelhead diets were dominated by invertebrates (*Bythotrephes* and Terrestrials, respectively) at small sizes and transitioned to much greater reliance on small and large alewives as their total length increased (Fig. 11 and Fig. 12). Brown trout were also largely reliant on alewives with a transition to larger alewives as total length increased (Fig. 13). Relative to other salmonines, Brown trout were more reliant on round goby as a prey source particularly in the Southeast (Fig. 13). Lake trout mixing model results suggest that they are primarily reliant on large alewives with secondary contributions from round goby (Fig. 14). This pattern appears to vary regionally, with greater reliance on round goby in the north part of the basin (Fig. 14). Notably less ontogenetic diet variation was observed in lake trout than other salmonines.

Long-term Trends

Analyses of historic stable isotope samples was focused on Chinook salmon, Coho salmon, and lake trout frozen tissue homogenates (Fig. 15). Results suggest that there has been little consistent temporal shift in δ^{13}C across locations, with values fluctuating between -24‰ and -22‰ for all three species. However, Chinook Salmon δ^{15}N did exhibit a consistent decline from 2000 to 2008 across study regions. In contrast, Coho Salmon and Lake Trout δ^{15}N did not exhibit consistent patterns temporally or spatially with respect to δ^{15}N.

Fatty Acid Results (Table 1c)

Fatty acids of prey species

Fatty acid profiles of prey items were relatively species-specific when assessed with linear discriminant analysis (Table 3). However, some species exhibited similar profiles, and there were some misclassification of profiles amongst them. For example, alewife and bloater had several individuals that were misclassified as the other species, suggesting similar feeding ecologies. Species that shared misclassifications also clustered closer together in nMDS plots (Fig. 17). In nMDS space alewife and round goby represented the ends of a gradient in fatty acid profiles, respectively representing pelagic and benthic components of food webs.

Dietary interpretations of salmon fatty acid profiles

We note that the ratio of 16:1n-7 to 18:1n-9 has been used to differentiate between alewife and round goby, and represents fatty acids that describe differences in their profiles as assessed by SIMPER (Table 4). We use this ratio to help us describe foraging patterns in the top predators of the system (Fig. 16). With this simple ratio we note that Lake Trout and Brown Trout have greater indicators of round goby consumption whereas the other salmonids have greater indicators of alewife consumption.

For Chinook Salmon and Lake Trout seasonal differences in fatty acid profiles were stronger than the regional differences (Table 5). As such, seasonal differences are more readily visible when profiles of each species are plotted compared to the regions (Fig. 19). These seasonal shifts likely correspond to greater diet diversity in the later seasons for Chinook Salmon, and more Round Goby in diets of Lake

Trout in the spring (Fig. 2). Although not shown, other species exhibited similar trends where seasonal differences were larger than regional ones.

For salmonids, fatty acid profiles appear to be affected by both the length of the individual and by the actual lipid content of the sample. We plotted each fatty acid profile in nMDS space and used average prey profiles as reference points (Fig. 18). Salmonid fatty acid profiles that shift more positively along the y-axis indicate a diet composition shift towards more Round Goby consumption. Concomitantly, fatty acid profile shifts along the x-axis indicate a shift due to lipid content changes in the individual salmon. As such, for Chinook or Coho Salmon it is apparent that there is only a shift with size that is due to lipid content changes, whereas for Brown Trout and Lake Trout there are strong shifts due to both a diet change and a lipid content change.

Conclusions

This project offers important insights into upper food web trophic structure and energy flow by providing high spatiotemporal resolution information regarding the diets of Lake Michigan salmonines. Recent piscivore diet studies in Lake Michigan have focused primarily on Chinook salmon and lake trout with less work on brown trout, Coho salmon, and steelhead. Prior to this study, the extent of nearshore resource use (e.g. consumption of round goby) by these species was unknown in Lake Michigan. This study fills this knowledge gap and provides the first evidence of how these species have adjusted their diets following establishment of dreissenid mussels and round goby in the Lake Michigan food web.

In addition to providing updated high spatiotemporal resolution diet data, this project generated the first estimates of niche overlap and diet proportion data based on stable isotopes for the Lake Michigan piscivore community. These data provide important insight into upper food web structure and potential interactions (e.g. competition) among species. In addition, the mixing model results provide the only known information on long-term integrated diet across regions and ontogenies in Lake Michigan piscivores. Establishing differences between observed diets and stable isotope-inferred diets facilitates a more mechanistic understanding of the benefits and biases associated with these approaches.

This project also represents one of the larger databases on fatty acid profiles at the top of a freshwater foodweb. For freshwater systems, knowledge of how fatty acids can be used to trace diets of wild predators comes from zooplankton and algae studies. Aquaculture studies have provided the ground work for proving that fatty acids of salmonines reflect their diets, but little is known about how wild-type diets (i.e., not formulated mixtures of ingredients) are reflected in the fatty acid profiles of salmonines. The combined analysis of stomach content and stable isotopes aides in defining specific trophic tracers within the fatty acid data set that can aide in future studies using this technique.

To date, the major findings of this project include:

1. Coho and Chinook rely heavily on alewife, and appear to rely less on round goby than in Lake Huron, possibly because alewife remain more abundant in lake Michigan.
2. Steelhead trout have a diverse diet, and rely to a significant extent on terrestrial insects at some times of year.
3. Lake trout and brown trout have diverse diets, allowing for reduced dietary overlap with other salmonines.
4. Large lake trout have high δ^{15}N values, possibly reflecting an increased dependence on profundal food sources.

5. Lake trout, brown trout and burbot rely quite heavily on round goby, with the dependence on round goby increasing with fish size.
6. Burbot appear to have reduced diet diversity over time, with an increased reliance on round goby as a food source. However sculpin remain an important food source for burbot in southern part of the lake.
7. In general, salmonines appear to feed more heavily on alewife on the west side of lake, and gobies on the east side.
8. Burbot and yellow perch rely heavily on nearshore energy sources.
9. Lake whitefish diet is highly spatially variable. Chironomids appear to be an important food source in the lake's northeast quadrant, while Mysis are important in the southeast quadrant. Stomach content analysis suggests they do not feed heavily on quagga mussels.
10. Regional differences in fatty acid composition of individual prey species was minimal. Therefore, the observed regional differences in the fatty acid profiles of some of the salmonines (especially lake trout) likely reflect real regional differences in diet, although the apparent effect of lipid content on fatty acid composition must be considered when interpreting regional and spatial differences.

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Grant No. 2015.1550

Project Title:

The New Lake Michigan Food Web:

Establishing links between nearshore food sources and pelagic piscivores

Grantee Organization: University of Wisconsin-Milwaukee

Project Team: Principle Investigator: Harvey Bootma (University of Wisconsin-Milwaukee); Co-investigators: Sergiusz Czesny (U. of Illinois), Tomas Hook (Purdue University), and Jacques Rinchard (SUNY-Brockport).

Contact Person: Harvey Bootsma, hbootsma@uwm.edu, 414-382-1717

Grant Amount: $264,772.30

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Key Search Words: Lake Michigan, food web, trophic, salmon, trout, round goby, stable isotopes, fatty acids, stomach content

Background/Overview:

1. Broad scale changes in nutrient-algal dynamics, energy flows, and relative species abundance have been relatively well documented in Lake Michigan in recent years. There have been declines in offshore pelagic productivity especially with respect to lower trophic levels and the loss of the spring diatom bloom, declines in Diporeia abundance, and declining preyfish biomass. Concurrent with loss of pelagic productivity and biomass has been an increase in nearshore production especially with respect to benthic algae and the invasive round goby. Uncertainty in how the pelagic piscivores have responded to declining pelagic energy sources by relying more on nearshore energy sources prompted a research need.

   To address this need, this project addressed two major objectives: a) **surveys to determine the spatial patterns of trophic structure and fish condition**, and b) **assessment of long-term trends in energy flow and fish condition in the upper food web**. To achieve these objectives, there are 5 major tasks: 1) collection of fish for regional comparisons of upper food web structure, 2) regional comparison of upper food web structure, 3) confirmation of spatial trends in nearshore trophic structure, 4) determine long-term trends in upper food web structure, and 5) analysis, presentation and publication of data.

2. Although all major project tasks were completed, several deviations from the original plan of work occurred because it was difficult to obtain the samples necessary to fully complete all tasks. A.) Limited numbers of lake whitefish, burbot and yellow perch were obtained for spatial comparison. Because these species are not collected by USFWS biotechnicians as part of their Mass Marking Program, collections were largely reliant on fishery independent surveys. These

surveys (e.g. Lakewide Assessment Plan, LWAP) yielded fewer samples with lower spatial resolution than was achieved through the USFWS Mass Marking fishery dependent sampling used to obtain most salmonine species. As a result, spatial comparisons related to Task 2, (regional comparison of upper food web structure) were largely limited to the salmonine complex. B.) Task 4, assessment of long-term trends in upper food web structure, required that piscivores stable isotopes be compared relative to a stable C and N isotopic baseline. Historic invertebrate and preyfish samples were not available to establish this baseline from which upper food web structure could be assessed. As a result, historic changes in stable C and N isotope ratios of several Lake Michigan piscivores were measured but conclusions regarding changes in upper food web structure await further information on long-term trends in baseline organisms. P.I. Harvey Bootsma has an archive of stable C and N isotope data from historic sediment samples provided by NOAA/GLERL which may offer an alternative baseline moving forward. We intend to analyze this external dataset within the next 6 months.

Outcomes:

3. This study advanced scientific knowledge related to upper food web structure and salmonine diets by:
    a. *Providing an examination of multiple salmonine species diets across the entire Lake Michigan Basin*. Although multiple studies of salmonine diets have been done in Lake Michigan, this study is the first to examine multiple species across the entire Lake Basin. This approached has facilitated interspecific and regional comparisons of diet differences which can be used to generate and inform hypotheses regarding the mechanisms for this variation (e.g. the importance of nearshore prey such as round goby as it relates to dominant substrate type).
    b. *Addressing diet composition using multiple trophic indicators*. This is the first study which has used gut contents, stable isotopes, and fatty acids trophic indicators to assess the diets of Lake Michigan salmonines. These three methods provide different levels of specificity for consumed prey, reflect feeding over different temporal scales ranging from hours (gut contents) to weeks (fatty acid analysis) to years (stable isotope analysis), and are complementary to one another (Alfaro et al. 2006).
    c. *Providing data on both predator and prey fatty acid profiles*. The use of fatty acids as trophic indicators is a technique that continues to be refined, especially in the realms of freshwater fishes. When combined with stable isotope and gut content data, insights into how factors such as ontogeny, lipid content, and diet composition affect a predator's fatty acid profile can be ascertained. Further, comparison of fatty acid profiles collected by this project to those collected over a decade ago offers an opportunity to explore temporal fatty acid profile dynamics.
    d. *Providing stable C and N isotope data to inform mixing models describing long-term regional diet patterns and ontogenetic diet differences*. Stable isotope analysis has been important tool for ecologists tracking energy and nutrient flows in food webs for decades, but recent advances in computational power and analytical tools offer new opportunities to quantitatively describe and compare diet composition and niche overlap with multiple prey sources. This study offers the first detailed stable isotope

comparison of regional and ontogenetic diet differences in the Lake Michigan salmonine complex.

4. This project provided funding to support a master's student (Ben Leonhardt) at Purdue University and a PhD student (Ben Turschak) at UW-Milwaukee. In addition to providing support for graduate research directly related to project objectives, multiple undergraduate and master's students from Purdue, UW-Milwaukee, and SUNY-Brockport assisted with sample collection, processing, and analysis wherein they were able to develop skills pertinent to Great Lakes fishery research.
5. Several new relationships and collaborations among team members and the research and management communities were develop over the course of this project which greatly aided in project completion.
    a. US Fish and Wildlife Service: The project team developed a strong partnership with USFWS, especially those involved with the Mass Marking Program. This relationship resulted in many of the samples needed for comparison of upper trophic level spatial patterns, especially with respect to the salmonine community. Additional support was provided by USFWS to collect non-salmonine piscivorous species including Lake Whitefish and Burbot. Project partners continue to work with USFWS collaborators to provide analytical support for USFWS-collected stable isotope samples from 2014.
    b. EPA: Collaboration with EPA Great Lakes National Program Office was critical to securing hundreds of historic frozen tissue homogenate samples from Lake Michigan salmonines for historic stable isotope analysis.
    c. Little Traverse Bay Band of Odawa Indians: LTBBOI provided many of the samples from the Northeastern region of Lake Michigan where it was difficult to secure enough samples from field collection or from other project collaborators. Project partners continue to work with LTBBOI collaborators to develop and implement a study of food web structure in northeast Lake Michigan using stable isotopes.
    d. Michigan and Wisconsin DNR: Michigan and Wisconsin DNR provided many preyfish and piscivore samples from around the Lake Michigan Basin for analysis of upper trophic level spatial patterns. In addition, both MI and WI DNR pledged historic scale samples for analysis of long-term trend in energy flow. Project partners continue to work with DNR collaborators to inform and update inputs to the Predator Prey Ratio Model (PPR Model) using results from this project.
    e. NOAA: NOAA Great Lakes Environmental Research Laboratory assisted greatly in providing forage fish from southeastern Lake Michigan for confirmation of spatial patterns in forage fish diet and source data for isotope mixing models. NOAA/GLERL has also provided a large, historical stable isotope data set going back to the 1970's, which we hope to use to determine trophic baseline isotope ratios with which historical fish samples can be compared.
6. Ongoing management effort on Lake Michigan focuses strongly on balancing predator biomass with available forage. Though several iterations of this approach have been used over time, most recent efforts rely on the ratio of Chinook salmon to Alewife biomass estimated using statistical catch at age (SCAA) models. The model for alewife is largely dependent on accurate

assessment of alewife consumption (i.e. the “catch” component of the model) by all major predators and relies on diet information from Stewart and Ibarra (1991) which are now several decades old. Members of the project team are playing an active role on the Salmonine Working Group of the Lake Michigan Technical Committee to update diet information with the data collected in this project. Future iterations of these models are being developed to include time-varying diet data (seasonal and interannual), spatial diet data, and additional predator and prey SCAA models based largely on the outcomes of this project. Results of these models will in turn directly impact management decisions such as stocking.

7. The most important outcome resulting from this project is the relatively high spatiotemporal resolution piscivore diet information using 3 complementary methods (i.e Gut Contents, Stable Isotopes, and Fatty Acids). While most recent studies have focused on a single species (e.g. Lake trout and Chinook Salmon), region, or method, this project provides much more comprehensive diet information. This is especially noteworthy with respect to lesser studied salmonines such as Coho salmon, Steelhead, and Brown Trout. Prior to this study, the extent of nearshore resource use/consumption of round goby by brown trout, Coho salmon, and steelhead in Lake Michigan was unknown. This study fills major diet gaps that were missing for these species and offers insight into how they have responded to major ecological changes. Furthermore, these updated diet data are critical to informing ongoing management efforts including the SCAA models described above.

Related Efforts:

8. Many of the samples provided for this project were collected as part of broader efforts in the management and research community of Lake Michigan. In particular, the samples collected as part of the USFWS Mass Marking program and fall assessments; Michigan and Wisconsin DNR Spring and Fall Lake Wide Assessments as well as commercial fisher sampling; GLERL bottom trawling, and LTBBOI gillnet surveys and commercial fisher sampling were critical to achieving necessary sample sizes. Furthermore, historical samples would not have been available without independent past efforts my EPA/GLNPO, Michigan and Wisconsin DNR’s, and NOAA/GLERL.
9. Several spin-off projects related to this project have been initiated. A) A study lead by P.I. Brian Roth from MSU titled, “A comparison of predator diets and stable isotopes in Lake Michigan and Huron” was recently approved for funding by Great Lakes Fisheries Trust (2018.1783); B) A study led by P.I. Jason Smith from LTBBOI to investigate trophic structure in northeastern Lake Michigan particularly as it relates to Cisco has been funded by BIA; and C) the Salmonine Working Group of the Lake Michigan Technical Committee is currently working on updating and developing predator and prey SCAA models with updated diet and consumption data from this project. Each of these spin-off projects was initiated and/or draws on information generated in this project. Furthermore, all spin-off projects include at least one member of the existing project team.

Communications/Publication of Findings:

Presentations

10. Presentations
    a. Bootsma. What's Happening to the Great Lake Food Chain? 2017 Michigan Charter Boat Association, Holland, MI (Oral Presentation)
    b. Kornis and Turschak. Salmon and Trout Diets in Lake Michigan.
        i. 2018 Sea Grant Anglers Workshop. Ludington, MI (Oral Presentation)
        ii. 2018 GLFC Upper Lakes Meeting, Sault Ste. Marie, ON (Oral Presentation)
    c. Leonhardt et al. Size- Specific Consumption by Lake Michigan Piscivores.
        i. 2017 Purdue University Forestry and Natural Resources Research Poster Competition, West Lafayette, IN (Poster Presentation)
        ii. 2017 IAGLR, Detroit, MI (Poster Presentation)
        iii. 2017 National AFS Meeting, Tampa, FL, (Oral Presentation)
    d. Leonhardt et al. Describing the Diet Complexity of Lake Michigan Salmonines. 2017 State of Lake Michigan Meeting, Green Bay, WI (Oral Presentation)
    e. Leonhardt et al. Changes in the Prey Consumption Patterns of Lake Michigan Salmonines. 2017-2018 Illinois-Indiana Sea Grant Fisheries Workshop, Michigan City, IN and Chicago, Illinois (Oral Presentation)
    f. Leonhardt et al. Diet Complexity of Lake Michigan Salmonines: Contrasting Trophic Indicators.
        i. 2018 Midwest Fish and Wildlife Conference, Milwaukee, WI (Oral Presentation)
        ii. 2018 Indiana AFS, Lafayette, IN (Oral Presentation)
        iii. 2018 IAGLR, Toronto, Canada (Oral Presentation)
    g. Turschak and Bootsma. Using Stable C and N isotopes to Characterize Large-Scale Spatial and Temporal Variation in the Diets of Lake Michigan Fishes. 2017 Midwest Fish and Wildlife Conference, Lincoln, NE (Oral Presentation).
    h. Turschak et al. Using Stable Isotopes to Assess Diets of Lake Michigan Salmonines: Implications for Ongoing Management. 2018 Midwest Fish and Wildlife Conference, Milwaukee, WI (Oral Presentation)
    i. Turschak. The Growing Importance of the Nearshore Zone. 2018 Michigan Sea Grant Anglers Workshop. Ludington, MI (Oral Presentation)
    j. Turschak et al. Effects of Ecology and Biogeochemistry on the Stable Isotopes of Nearshore Fishes in Lake Michigan. 2017 IAGLR, Detroit, MI (oral Presentation)
    k. Bunnell, D.B., R. Barbiero, H. Bootsma, H. Carrick, R. Claramunt, J. Dettmers, A. Elgin, Y.-C. Kao, B. Lesht, B. Hinchey-Malloy, C. Madenjian, K. Pangle, S. Pothoven, C. Riseng, M. Rowe, E. Rutherford, S. Thomas, B. Turschak, H. Vanderploeg, D. Warner. Exploring how lower trophic level changes influence prey fish in Lake Michigan. Presentation given to the Great Lakes Fishery Commission, March 22, 2017.
    l. Feiner, Z. C. Foley, R. Swihart, H. Bootsma, S. Czesny, J. Janssen, J. Rinchard, T. Höök. Species-specific spatial patterns of trophic complexity in a Lake Michigan food web. State of Lake Michigan Conference, Green Bay, WI, Nov. 7-10, 2017.

Publications

11. Publications

    a. Kornis and Turschak. What does a changing forage base mean for Lake Michigan Salmon and Trout. 2018 Great Lakes Sport Fishing News.
    b. Leonhardt et al. What's on the Menu for salmon and trout in Lake Michigan? In Press. Illinois-Indiana Sea Grant Extension Publication
    c. Leonhardt, Benjamin. (2018). MS Thesis. Evaluating Methods to Describe Dietary Patterns of Lake Michigan Salmonines. Purdue University, West Lafayette, Indiana, USA.

12. Project results and updates have been regularly shared with the Lake Michigan Technical Committee. This included presentation of gut content data at the July 2017 LMTC meeting in Kenosha, WI; presentation of stable isotope and gut content data at the January 2018 LMTC meeting in Michigan City, IN; and presentation of fatty acid, stable isotope and gut content data at the July 2018 LMTC meeting in Manistee, MI. These data were also shared with the Great Lake Fishery Commission as well as other tribal, management, and research agencies at the March 2018 Upper Lakes Meeting in Sault Ste. Marie, ON. In addition to sharing with research and management agencies, results have also been shared with stakeholder groups including at the 2017 Lake Michigan Charter Boat Association Meeting in Holland, MI and the 2018 Sea Grant Anglers Workshop in Ludington, MI.

Discussion

Stomach Contents

Despite salmonine species having distinct feeding patterns, alewife were clearly the dominant prey item in the stomachs of Lake Michigan salmonines (Fig. 1-3). This was particularly true for Chinook salmon, which almost exclusively consumed alewife and had little contribution from other prey items like bloater, round goby, yellow perch, and invertebrates. Our observation of Chinook salmon feeding almost exclusively on alewife (proportional diet composition: 73%; mean g/stomach: 9.0 g; Fig. 1 -3) despite depressed alewife populations is consistent with previous studies in lakes Michigan (Jacobs et al. 2013) and Huron (Roseman et al. 2014). Based on the percent diet composition by weight, Coho salmon primarily consumed alewife and aquatic invertebrates (*Bythotrephes* and *Mysis*), but the mean weight of alewife in Coho salmon stomachs was nearly three times the weight of aquatic invertebrates (Fig. 1). This might suggest that Coho salmon are acquiring the bulk of their energy from alewife despite the appearance of a diverse diet composition. Steelhead were different from other salmonine species in that terrestrial insects contributed significantly to their stomach contents (Fig. 1-3). This diet pattern is consistent with steelhead in Lake Huron where terrestrial insects were the dominant prey item following declines in alewife abundance (Roseman et al. 2014). It is likely that some biases associated with stomach content analysis (longer digestive rates for terrestrial insects compared to soft-bodied prey; Kionka and Windell 1972) and angler-caught fish (anglers targeting steelhead at thermal bars where both steelhead and terrestrial insects accumulate; Aultman and Haynes 1993, Höök et al. 2004, Roseman et al. 2014) could have affected our results.

Of the salmonine species, the primary consumers of round goby were brown trout and lake trout (Fig. 1-3). Our observations are consistent with previous work in lakes Michigan and Huron, showing that lake trout have increased their reliance on round goby following the decline in alewife abundance (Happel et al., 2017; Jacobs et al., 2010; Roseman et al. 2014). Additionally, brown trout and lake trout in Lake Ontario appear to consume more round goby than the other three species (Yuille et al. 2015, Happel et al. 2016). Although stomach contents revealed that Chinook salmon, Coho salmon, and steelhead consume round goby, round goby contributed minimally to their diet composition. It was somewhat surprising that round goby did not contribute more to the diets of Coho salmon and steelhead, since round goby make up roughly 10-15% of Coho salmon and steelhead by weight in Lake Huron (Roseman et al. 2014). This might suggest that alewife abundance in Lake Michigan may be sufficiently high enough to allow Coho salmon and steelhead to continue to feed heavily on alewife rather than other fish prey, like round goby. Lastly consistent with our stomach content findings, there has been little evidence of Chinook salmon feeding on round goby in lakes Huron (Roseman et al. 2014) and Ontario (Yuille et al. 2015, Happel et al. 2016), which shows the potential diet inflexibility of Chinook salmon in the Laurentian Great Lakes.

Stomach contents revealed spatio-temporal feedings patterns of Lake Michigan salmonines, particularly in the spring (Fig 2). On the western side of Lake Michigan in the spring, all five salmonine species almost exclusively consumed alewife. Previous research has shown higher densities of alewife on the western side of Lake Michigan in the spring (Brandt et al. 1991). High densities of alewife on the western side of the lake may have made it unnecessary to feed on other prey items unlike on the eastern side of Lake Michigan where salmonines had more diverse diet compositions. Additionally, we observed increased round goby consumption by brown trout and lake trout on the eastern side of Lake Michigan in the spring (Fig. 2). This pattern has been previously observed in the stomach contents of lake trout collected from Lake Michigan in the spring of 2011 by Happel et al (2017). The western shoreline of Lake Michigan has much more complex, rocky habitat compared to the eastern shoreline (Foley et al., 2017; Happel et al., 2015a, b), which may decrease the availability and increase handling time of round goby compared to the eastern side. Additionally, the combination of round gobies moving offshore to overwinter (Kornis et al. 2012) and salmonines occupying nearshore areas in the spring (Olson et al. 1988) may increase interactions between round goby and brown trout and lake trout.

The stomach contents of burbot revealed that they are feeding on round goby, which is consistent with studies in lakes Michigan, Huron, and Erie (Fig. 6; Hensler et al. 2008, Stapanian et al. 2011, Hares et al. 2015)). Past studies revealed that burbot in Lake Michigan have a diverse diet, which included a mixture of alewife, sculpins, round goby, sticklebacks, bloaters, and crayfish (Hares et al., 2015; Hensler et al., 2015). Stomach contents of burbot collected in 2016 and 2017 suggest that burbot have switched from consuming a diverse diet to almost exclusively consuming round goby (Fig. 6). Although other prey items contributed little to the lake wide diet compositions in 2016 and 2017, sculpins contributed relatively large proportions in the southern regions of Lake Michigan, which is consistent with past work (Hares et al., 2015).

In the stomachs of lake whitefish, we observed strong spatial patterns in the diet composition of lake whitefish (Fig. 7). The spatial patterns observed in 2016 stomach contents are generally consistent with recent lake whitefish diet work completed in Lake Michigan(Pothoven and Madenjian 2008) with some exceptions. Past work indicated that *Diporeia* were an important diet component (13-15%; Pothoven and Madenjian, 2008), but *Diporeia* were not observed in stomachs of lake whitefish collected in 2016. This is likely attributed to the observed declines in *Diporeia* abundance in Lake Michigan (Nalepa et al., 2009). Additionally, Pothoven and Madenjian (2008) documented that dreissenid mussels contributed significantly to the diet composition (26-42%) of lake whitefish in northeast and southeast regions of Lake Michigan, which is inconsistent with our observations. Of all the lake whitefish stomachs collected and analyzed in 2016, dreissenid mussels were found in only 8 stomachs. It is unclear if this can be attributed to yearly variation in feeding patterns of lake whitefish or if lake whitefish are relying less on dreissenid mussels compared to the past.

Stable Isotopes

Stable C and N isotope ratios of most Lake Michigan salmonines indicated that they occupy a relatively similar trophic role (Fig. 8). Lake wide mean δ^{13}C, which provides an indication of primary energy source (pelagic vs. benthic; France 1995, Hecky and Hesslein 1995), suggests that these species all are highly reliant on pelagic energy sources relative to other piscivores such as burbot and yellow perch that appear to be much more reliant on nearshore energy sources (Fig. 8). Mean δ^{15}N, which is an indicator of trophic level and/or pelagic vs profundal feeding (Minagawa and Wada 1984, Sierszen et al. 2014), indicates that all salmonines occupy the upper pelagic food web except Lake Trout which had a much greater mean δ^{15}N (Fig. 8 and Fig. 9). The elevated δ^{15}N of Lake Trout suggests that they are more reliant on profundal energy sources than other salmonines (Sierszen et al. 2014). Relative to salmonines, burbot and large yellow perch had a much heavier δ^{13}C indicating greater reliance on benthic or nearshore prey sources (France 1995, Hecky and Hesslein 1995) however, yellow perch appear to have a lower trophic level than other piscivores which may be indicative of greater reliance in invertebrate prey (Turschak and Bootsma 2015, Turschak et al. 2018).

To better elucidate trophic differences among Lake Michigan salmonines, isotopic niche overlap was measured by fitting 95% Bayesian ellipses to the data and calculating overlap among the species (Table 2, Fig 8; Jackson et al. 2011, Swanson et al. 2015). Lake Trout and brown trout had the largest niche area indicating increase diet flexibility or regional variation. These broader niche areas also corresponded to lower probability of niche overlap with other salmonines. Increased niche area and apparent diet flexibility among these species is likely related to increased reliance on round goby as a prey item (Table 2, Fig. 1 and Fig. 9). Other studies have also shown greater diet diversity in these species although alewives are still a dominant prey item in their diets (Dietrich et al. 2006, Mumby et al. 2018). Steelhead occupied an intermediate niche area and spanned a broader δ^{15}N range which likely demonstrates a flexible pelagic foraging behavior that includes terrestrial invertebrates as well as prey fish such as alewife (Fig. 9; Aultman and Haynes 1993, Rand et al. 1993, Roseman et al. 2014). By comparison, Chinook salmon occupied a smaller niche area and had high probability of niche overlap with other salmonines. This is likely related to very high reliance on Alewife prey (Table 2, Fig. 1 and Fig. 9). Coho salmon occupied the smallest niche area with an intermediate probability of overlap with other salmonines. This is likely related to their lower overall trophic position and reliance on invertebrate prey and smaller alewives (Roseman et al. 2014). A recent study of salmonine isotopic niche overlap in Lake Ontario revealed similar findings with the lowest niche overlap observed in Lake Trout and Brown trout and highest overlap observed in Chinook and Coho Salmon (Mumby et al. 2018). The authors also attributed the high degree of niche overlap in Chinook and Coho salmon to reliance on alewife prey. Similarity between these findings again highlights the inflexibility of Chinook and Coho Salmon to change their diet despite declines in their preferred forage (Jacobs et al. 2013).

Stable isotope mixing model results corresponded well with stomach content data (Mean Percent by Weight). Observed diet generally fell within the estimated Bayesian confidence intervals and adds credibility to project results (Fig. 1, Fig. 3, and Fig. 10-14). However, stable isotopes mixing models indicated several notable differences including: 1) the higher proportion of bloater in Chinook Salmon diets, 2) higher proportions of invertebrate in Coho Salmon and Steelhead diets, and 3) smaller regional differences in diet proportions. Apparent differences between these methods likely results from several potential factors. In particular, stable isotopes integrate over a much longer time scale and as a result provide a view of diet patterns over months to years (Vander Zanden et al. 1997, Newsome et al. 2007). Gut contents by comparison offer a snap shot of diets and are subsequently more subject to biases associated with fishery dependent sampling (Aultman and Haynes 1993, Höök et al. 2004, Roseman et al. 2014). However, gut contents provide much greater taxonomic resolution especially when prey sources are isotopically similar as was observed for alewives and bloater in this study (Parnell et al. 2010). This isotopic similarity in prey source data led to less certainty in mixing model results for some species (Parnell et al. 2010).

Analyses of historical stable isotope samples was impeded by small sample sizes and data gaps. However, consistent declines in Chinook Salmon δ^{15}N may be attributable to reduced availability of larger alewives or shifts in the isotopic baseline (Warner et al. 2008, Turschak et al. 2014). Coho Salmon and Lake Trout δ^{15}N were more variable temporally and less consistent across regions. Mechanisms driving temporal isotopic variation are difficult to discern without knowledge of isotopic baseline (Vander Zanden and Rasmussen 1999, Post 2002). However, given the lack of consistency across the time series, regions, and species, it does not appear that increased nearshore energy availability has affected pelagic predators in the same way as it has other fishes and invertebrates (Turschak et al. 2014).

Fatty Acids

A ratio of 18:1n-9 (Oleic acid;OA) to 16:1n-7 (Palmitoleic acid; POA) was suggested as a means of tracing pelagic (i.e., Alewife) vs benthic/littoral (i.e., Round Goby) foraging (Happel et al. 2017b). As such, these ratios were calculated and explored to assess each species' reliance on the two prey species (Fig. 16). We note that Brown Trout and Lake Trout trend slightly more towards indicators of Round Goby (higher 16:1n-7) than other species analyzed. This corresponds to higher Round Goby masses in Lake Trout and Brown Trout stomach contents. Also, there seems to be more 16:1n-7 in Steelhead caught in the Eastern regions of the lake than in the Western Regions. This could either be due to a greater reliance on Alewife in western regions of the lake compared to eastern regions where diets were more varied.

Classification of each prey sample during LDA indicated that the fatty acid profile of each prey species was relatively distinct (Table 3). Some ecologically similar species had rather similar fatty acid profiles, for example the misclassifications among Alewife and Bloater. Prey sample fatty acid profiles were visualized using nMDS plots, and vectors used to assess which fatty acids can be used to distinguish between prey species (Fig. 17). Few regional differences in fatty acid compositions within each prey species were noted in nMDS space. Differences among species were primarily along the y-axis of the nMDS plot. Fatty acids that correlated positively along this axis, indicative of Round Goby, included those of the n-7 family, 22:5n-3, and 20:5n-3 (EPA). Conversely fatty acids that strongly correlated negatively and thus are more indicative of Alewife included 22:6n-3 (DHA) and several polyunsaturated n-3 fatty acids. SIMPER analysis of the differences between Alewife and Round Goby supported our interpretations of the vector analysis: EPA and n-7 fatty acids were higher in Round Goby whereas n-3 and n-9 fatty acids were higher in Alewife (Table 4).

We maintained Alewife, Bloater, Rainbow Smelt, and Round Goby nMDS coordinates and summarized them as means (95% CI) to illustrate differences in salmonine fatty acid profiles (Fig. 18). Each of the 5 main salmon of interest was plotted separately with a color gradient used to show how fatty acids shift with the length of the individual. Further investigation indicated that points spreading from left to right along the x-axis were shifting (i.e., correlated) with lipid content (increasing proportions of 18:1n-9). Conversely, points that shifted from the bottom up were shifting along a length gradient more than a lipid gradient. As such, within a salmon species, a bottom up shift likely corresponds to an ontogenetic diet shift from Alewife to Round Goby. Happel et al. (2018) showed that larger Lake Trout are more likely to consume Round Goby than smaller individuals. Our data herein suggests a similar diet shift occurs with Brown Trout whereas Chinook and Coho Salmon appear not to stray from Alewife consumption.

LDA analysis suggest that season of capture and region of capture affected salmon fatty acid profiles (Table 5). All species had significant effects of Region and Season when assessed with PERMANOVA ($P < 0.05$). While there does appear to be some misclassification among regions for Chinook Salmon and Lake Trout there appears to be little misclassification between seasons. This would indicate that there is a stronger shift in fatty acid profiles between seasons than between regions. We do note that within the Lake Trout fatty acid profiles, those caught in the North East region from the Spring are the most distinct (Fig. 19). This is primarily driven by high 22:6n-3 and 18:0 content in these lake trout. However, it remains to be assessed if this shift is due to differences in length and lipid content or due to actual changes in foraging habits.

Benefits, Challenges, Surprises and Lessons Learned

Primary benefits of this work include updating diet and consumption estimates of predators which will improve SCAA models of forage fish biomass. Researchers and managers—facing a changing forage base and reduced alewife abundance in Lake Michigan—are eager to incorporate these findings into these models which are the primary management tool used for determining appropriate stocking levels in Lake Michigan.

Though this work has important benefits, several major challenges had to be overcome to reach this point. In particular, project progress at times was impeded by equipment failures including the IR-MS and GC MS instruments needed to perform stable isotope and fatty acid analyses. Additional challenges included low sample sizes for non-salmonine piscivores and difficultly establishing a historical baseline for assessment of long-term trends in upper food web energy flow.

Collaborations with other agencies were critical to gathering samples needed for this work. Without the help from the groups outlined above (see no. 5), much less data would have been available, and conclusions would have been severely limited. In addition to the exchange of samples and data, collaboration with these groups has resulted in widespread dissemination and application of project results. Although managing data and samples from multiple partners was logistically challenging, this allowed us to acquire a large number of samples from many regions of the lake, a task that would have not been possible had we implemented this project on our own. The new insights gained from this project are due in part to the multiple analytical methods used, but also to the extensive spatial and temporal coverage that was facilitated through a multi-agency collaborative approach.

References

Aultman, D.C., and Haynes, J.M. 1993. Spring thermal fronts and salmonine sport catches in Lake Ontario. North Am. J. Fish. Manag. **13**(3): 502-510.

Bootsma, H., Hecky, R.E., Hesslein, R.H., and Turner, G.F. 1996. Food partitioning among Lake Malawi nearshore fishes as revealed by stable isotope analysis. Ecology **77**: 1286–1290.

Brandt, S.B., Mason, D.M., Patrick, E.V., Argyle, R.L., Wells, L., Unger, P.A., and Stewart, D.J. 1991. Acoustic Measures of the Abundance and Size of Pelagic Planktivores in Lake Michigan. Can. J. Fish. Aquat. Sci. **48**(5): 894–908. doi:10.1139/f91-106.

Dietrich, J.P., Morrison, B.J., and Hoyle, J.A. 2006. Alternative Ecological Pathways in the Eastern Lake Ontario Food Web--Round Goby in the Diet of Lake Trout. J. Gt. Lakes Res. **32**: 395–400.

Dub, J. D. & Czesny, S. J. (2016). Yellow perch population assessment in southwestern Lake Michigan, Indiana Natural History Survey Technical Report 2015 (43).

Elliott, R. F., P. J. Peeters, M. P. Ebener, R. W. Rybicki, P. J. Schneeberger, R. J. Hess, J. T. Francis, G. W. Eck, and C. P. Madenjian. 1996. Conducting diet studies of Lake Michigan piscivores: a protocol. U.S. Fish and Wildlife Service, Report 96-3, Ann Arbor, Michigan.

Foley, C.J., Henebry, M.L., Happel, A., Bootsma, H.A., Czesny, S.J., Janssen, J., Jude, D.J., Rinchard, J., and Höök, T.O. 2017. Patterns of integration of invasive round goby (Neogobius melanostomus) into a nearshore freshwater food web. Food Webs **10**: 26–38. Elsevier Inc. doi:10.1016/j.fooweb.2016.10.001.

France, R.L. 1995. Carbon-13 enrichment in benthic compared to planktonic algae: foodweb implications. Mar. Ecol. Prog. Ser. **124**: 307–312.

Happel, A., Creque, S., Rinchard, J., Höök, T., Bootsma, H., Janssen, J., Jude, D., and Czesny, S. 2015a. Exploring yellow perch diets in Lake Michigan through stomach content, fatty acids, and stable isotope ratios. J. Great Lakes Res. **41**: 172–178. International Association for Great Lakes Research. doi:10.1016/j.jglr.2015.03.025.

Happel, A., Jonas, J.L., McKenna, P.R., Rinchard, J., He, J.X., and Czesny, S.J. 2017. Spatial Variability of Lake Trout Diets in Lakes Huron and Michigan Revealed by Stomach Content and Fatty Acid Profiles. Can. J. Fish. Aquat. Sci.: 1–11. doi:10.1139/cjfas-2016-0202.

Happel, A., Lafountain, J., Creque, S., Rinchard, J., Höök, T., Bootsma, H., Janssen, J., Jude, D., and Czesny, S. 2015b. Spatio-temporal description of spottail shiner (*Notropis hudsonius*) fatty acid profiles in Lake Michigan's southern basin. J. Great Lakes Res. **41**: 179–184. International Association for Great Lakes Research. doi:10.1016/j.jglr.2015.04.013.

Happel, A., Pattridge, R., Walsh, M., and Rinchard, J. 2016. Assessing diet compositions of Lake Ontario predators using fatty acid profiles of prey fishes. J. Great Lakes Res. International Association for Great Lakes Research. doi:10.1016/j.jglr.2016.12.008.

Hares, C.J., Jonas, J.L., and Leonard, J.B.K. 2015. Diet analysis of burbot (Lota lota) from eastern Lake Michigan: 1996–2012. Hydrobiologia **757**(1): 89–99. Springer International Publishing. doi:10.1007/s10750-015-2297-y.

Hecky, R.E., and Hesslein, R.H. 1995. Contributions of benthic algae to lake food webs as revealed by stable isotope analysis. J. North Am. Benthol. Soc. **14**(4): 631. doi:10.2307/1467546.

Hensler, S.R., Jude, D.J., and He, J. 2008. Burbot growth and diets in Lakes Michigan and Huron: an ongoing shift from native species to round gobies. Am. Fish. Soc. Symp. **59**: 91–110.

Höök, T.O., Rutherford, E.S., Brines, S.J., Schwab, D.J., and McCormick, M.J. 2004. Relationship between Surface Water Temperature and Steelhead Distributions in Lake Michigan. North Am. J. Fish. Manag. **24**(1): 211–221. doi:10.1577/M02-159.

Jackson, A.L., Inger, R., Parnell, A.C., and Bearhop, S. 2011. Comparing isotopic niche widths among and within communities: SIBER - Stable Isotope Bayesian Ellipses in R. J. Anim. Ecol. **80**(3): 595–602. doi:10.1111/j.1365-2656.2011.01806.x.

Jacobs, G.R., Madenjian, C.P., Bunnell, D.B., and Holuszko, J.D. 2010. Diet of lake trout and burbot in Northern Lake Michigan during spring: Evidence of ecological interaction. J. Gt. Lakes Res. **36**: 312–317.

Jacobs, G.R., Madenjian, C.P., Bunnell, D.B., Warner, D.M., and Claramunt, R.M. 2013. Chinook salmon foraging patterns in a changing Lake Michigan. Trans. Am. Fish. Soc. **142**(2): 362–372.

Kionka, B.C., and Windell, J.T. 1972. Differential movement of digestible and indigestible food fractions in steelhead, Salmo gairdneri. Trans. Am. Fish. Soc. **101**(1): 112–115.

Knight, R. L., Margraf, F. J., & Carline, R. F. (1984). Piscivory by walleyes and yellow perch in western Lake Erie. *Transactions of the American Fisheries Society*, *113*(January), 667–693. https://doi.org/10.1577/1548-8659(1984)113<677

Kornis, M.S., Mercado-Silva, N., and vander Zanden, M.J. 2012. Twenty years of invasion: A review of round goby Neogobius melanostomus biology, spread and ecological implications. doi:10.1111/j.1095-8649.2011.03157.x.

Minagawa, M., and Wada, E. 1984. Stepwise enrichment of d15N along food chains: Further evidence and the relation between d15N and animal age. Geochim. Cosmochim. Acta **48**: 1135–1140.

Mumby, J.A., Larocque, S.M., Johnson, T.B., Stewart, T.J., Fitzsimons, J.D., Weidel, B.C., Walsh, M.G., Lantry, J.R., Yuille, M.J., and Fisk, A.T. 2018. Diet and trophic niche space and overlap of Lake Ontario salmonine species using stable isotopes and stomach contents. J. Great Lakes Res. Elsevier. doi:10.1016/J.JGLR.2018.08.009.

Nalepa, T.F., Fanslow, D.L., and Lang, G.A. 2009. Transformation of the offshore benthic community in Lake Michigan: recent shift from the native amphipod Diporeia spp. to the invasive mussel Dreissena rostriformis bugensis. Freshw. Biol. **54**: 466–479.

Newsome, S.D., Rio, Martinez del, C., Bearhop, S., and Phillips, D.L. 2007. A Niche for Isotope Ecology. Front. Ecol. Environ. **5**(8): 429–436. doi:10.1890/060150.01.

Ngochera, M.J., and Bootsma, H.A. 2011. Temporal trends of phytoplankton and zooplankton stable isotope composition in tropical Lake Malawi. J. Gt. Lakes Res. **37**: 45–53.

Olson, R.A., Winter, J.D., Nettles, D.C., Haynes, J.M., Olson, R.A., Winter, J.D., Nettles, D.C., Nettles, D.C., and Haynes, J.M. 1988. Resource Partitioning in Summer by Salmonines in South-Central Lake Ontario. Trans. Am. Fish. Soc. **117**(6): 552–559. doi:10.1577/1548-8659(1988)117<0552.

Parnell, A.C., Bearhop, I.R., and JAckson, A.L. 2010. Source partitioning using stable isotopes: coping with too much variation. PLoS One **5**(3): e9672.

Post, D.M. 2002. Using stable isotopes to estimate trophic position: Models, methods, and assumptions. Ecology **83**(3): 703–718. doi:10.2307/3071875.

Pothoven, S. a., and Madenjian, C.P. 2008. Changes in Consumption by Alewives and Lake Whitefish after Dreissenid Mussel Invasions in Lakes Michigan and Huron. North Am. J. Fish. Manag. **28**: 308–320. doi:10.1577/M07-022.1.

Rand, P.S., Stewart, D.J., Seelbach, P.W., Jones, M.L., and Wedge, L.R. 1993. Modeling Steelhead Population Energetics in Lakes Michigan and Ontario. Trans. Am. Fish. Soc. **122**(5): 977–1001. Taylor & Francis Group. doi:10.1577/1548-8659(1993)122<0977:MSPEIL>2.3.CO;2.

Roseman, E.F., Schaeffer, J.S., Bright, E., and Fielder, D.G. 2014. Angler-Caught Piscivore Diets Reflect Fish Community Changes in Lake Huron. Trans. Am. Fish. Soc. **143**(6): 1419–1433. Taylor & Francis. doi:10.1080/00028487.2014.945659.

Sierszen, M.E., Hrabik, T.R., Stockwell, J.D., Cotter, A.M., Hoffman, J.C., and Yule, D.L. 2014. Depth gradients in food-web processes linking habitats in large lakes: Lake Superior as an exemplar ecosystem. Freshw. Biol. **59**(10): 2122–2136. doi:10.1111/fwb.12415.

Stapanian, M.A., Edwards, W.H., and Witzel, L.D. 2011. Recent changes in burbot growth in Lake Erie. J. Appl. Ichthyol. **27**(SUPPL. 1): 57–64. doi:10.1111/j.1439-0426.2011.01845.x.

Stewart, D.J., and Ibarra, M. 1991. Predation and production by salmonine fishes in Lake Michigan. Can. J. Fish. Aquat. Sci. **48**: 909–922.

Swanson, H.K., Lysy, M., Power, M., Stasko, A.D., Johnson, J.D., and Reist, J.D. 2015. A new probabilistic method for quantifying *n* -dimensional ecological niches and niche overlap. Ecology **96**(2): 318–324. Wiley-Blackwell. doi:10.1890/14-0235.1.

Traynor, D., Moerke, A., & Greil, R. (2010). Identification of Michigan fishes using cleithra. *Great Lakes Fishery Commission Miscellaneous Publication.* 2010-02.

Turschak, B.A., and Bootsma, H.A. 2015. Lake Michigan trophic structure as revealed by stable C and N isotopes. J. Great Lakes Res. **41**: 185–196. International Association for Great Lakes Research. doi:10.1016/j.jglr.2015.04.004.

Turschak, B.A., Bunnell, D.B., Czesny, S.J., Hook, T.O., Janssen, J., Warner, D.M., and Bootsma, H.A. 2014. Nearshore energy subsidies support Lake Michigan fishes and invertebrates following major changes in food web structure. Ecology **95**: 1243–1252.

Turschak, B.A., Czesny, S.J., Doll, J.C., Grunert, B.K., Hook, T.O., Janssen, J., and Bootsma, H.A. 2018. Spatial Variation in Trophic Structure of Nearshore Fishes in Lake Michigan as it Relates to Water Clarity. Can J. Fish. Aquat. Sci. **In Press**: 0.

Warner, D.M., Kiley, C.S., Claramunt, R.M., and Clapp, D.F. 2008. The Influence of Alewife Year-Class Strength on Prey Selection and Abundance of Age-1 Chinook Salmon in Lake Michigan. Trans. Am. Fish. Soc. **137**(6): 1683–1700. doi:10.1577/T07-130.1.

Yuille, M.J., Fisk, A.T., Stewart, T., and Johnson, T.B. 2015. Evaluation of Lake Ontario salmonine niche space overlap using stable isotopes. J. Great Lakes Res. **41**(3): 934–940. Elsevier.

doi:10.1016/J.JGLR.2015.05.011.

Vander Zanden, M.J., Cabana, G., and Rasmussen, J.B. 1997. Comparing trophic position of freshwater fish calculated using stable nitrogen isotope ratios (d15N) and literature dietary data. Can J. Fish. Aquat. Sci. **54**: 1142–1158.

Vander Zanden, M.J., and Rasmussen, J.B. 1999. Primary Consumer 13 C and 15 N and the Trophic Position of Aquatic Consumers. Ecology **80**(4): 1395–1404. doi:10.1890/0012-9658(1999)080[1395:PCCANA]2.0.CO;2.

Tables

Table 1a. Sample sizes for stomach content analyses.

	Early				Late				Grand Total
	NE	NW	SE	SW	NE	NW	SE	SW	
Brown Trout	**9**	**16**	**33**	**12**	**2**	**15**	**2**	**7**	**96**
<400		2	4	1	1			1	9
>400	9	14	29	11	1	15	2	6	87
Burbot	**18**		**14**		**8**	**10**		**3**	**53**
>300	18		14		8	10		3	53
Chinook Salmon	**5**	**45**	**72**	**48**	**30**	**45**	**35**	**25**	**305**
<600	4	23	34	26	15	23	16	7	148
>600	1	22	38	22	15	22	19	18	157
Coho Salmon	**5**	**24**	**68**	**41**	**27**	**3**	**33**	**25**	**226**
<600		8	42	19	18	1	8	13	109
>600	5	16	26	22	9	2	25	12	117
Lake Trout	**94**	**31**	**81**	**113**	**88**	**22**	**46**	**31**	**506**
<600	48	11	43	13	47	5	12	4	183
>600	46	20	38	100	41	17	34	27	318
Lake Whitefish	**11**		**20**			**27**			**58**
>300	11		20			27			58
Steelhead	**15**	**45**	**61**	**41**	**8**	**24**	**26**	**28**	**248**
<400		1				1			2
>400	15	44	61	41	8	23	26	28	244
Grand Total	**157**	**161**	**349**	**255**	**163**	**146**	**142**	**119**	**1492**

Table 1b. Sample sizes for stable isotope analyses.

	Early				Late				Grand Total
	NE	NW	SE	SW	NE	NW	SE	SW	
Alewife		**10**		**10**			**7**		**27**
<100		5		5			5		15
>100		5		5			2		12
Bloater		**5**		**5**	**2**	**2**			**14**
Brown Trout	**6**	**9**	**9**	**6**	**2**	**7**	**3**	**6**	**48**
<400		2	3	1	1		1	1	9
>400	6	7	6	5	1	7	2	5	39
Burbot	**5**		**5**		**6**	**5**		**4**	**25**
>300	5		5		6	5		4	25
Chinook Salmon	**5**	**11**	**10**	**12**	**11**	**11**	**11**	**11**	**82**
<600	4	5	5	6	5	6	6	6	43
>600	1	6	5	6	6	5	5	5	39
Coho Salmon	**5**	**10**	**10**	**8**	**10**	**3**	**11**	**10**	**67**
<600		5	5	3	5	1	5	5	29
>600	5	5	5	5	5	2	6	5	38
Deepwater Sculpin		**5**	**5**	**5**					**15**
Lake Trout	**6**	**8**	**10**	**10**	**4**	**10**	**12**	**10**	**70**
<600	5	3	5	5	4	5	6	5	38
>600	1	5	5	5		5	6	5	32
Cisco	**20**				**20**				**40**
<300	5								5
>300	15				20				35
Lake Whitefish	**5**		**5**			**5**			**15**
>300	5		5			5			15
Ninespine Stickleback		**5**							**5**
Rainbow Smelt		**5**		**5**		**3**			**13**
Steelhead	**5**	**6**	**5**	**5**	**5**	**6**	**6**	**5**	**43**
<400		1				1			2
>400	5	5	5	5	5	5	6	5	41
Round Goby	**14**	**10**	**15**	**10**	**15**	**2**	**13**		**79**
<60	4		5	5	5	1	5		25
>100	5	5	5		5	1	3		24
60-100	5	5	5	5	5		5		30
Slimy Sculpin		**1**	**5**	**5**					**11**
Spottail Shiner			**1**	**2**			**5**		**8**
Yellow Perch	**7**		**10**	**5**	**11**	**1**	**2**	**5**	**41**
<200	2		5	5	1	1	2		16
>200	5		5		10			5	25
Grand Total	**78**	**85**	**90**	**88**	**86**	**55**	**70**	**51**	**603**

Table 1c. Sample sizes for fatty acid analyses.

	Early				Late				Grand Total
	NE	NW	SE	SW	NE	NW	SE	SW	
Alewife		**10**		**10**			**6**		**26**
<100		5		5			5		15
>100		5		5			1		11
Bloater		**5**		**4**	**1**	**2**			**12**
Brown Trout	**8**	**15**	**19**	**12**	**2**	**15**	**2**	**7**	**80**
<400		2	4	1	1			1	9
>400	8	13	15	11	1	15	2	6	71
Burbot	**15**		**14**		**5**	**9**		**4**	**47**
>300	15		14		5	9		4	47
Chinook Salmon	**4**	**28**	**27**	**27**	**27**	**29**	**28**	**20**	**190**
<600	3	13	12	13	15	14	13	6	89
>600	1	15	15	14	12	15	15	14	101
Coho Salmon	**4**	**21**	**17**	**26**	**22**	**2**	**24**	**23**	**139**
<600		6	6	11	15	1	9	12	60
>600	4	15	11	15	7	1	15	11	79
Deepwater Sculpin		**5**	**5**	**5**					**15**
Lake Trout	**15**	**13**	**29**	**18**	**4**	**19**	**29**	**17**	**144**
<600	14	3	14	5	4	5	14	4	63
>600	1	10	15	13		14	15	13	81
Lake Whitefish	**12**		**16**			**15**			**43**
>300	12		16			15			43
Nine-Spine Stickleback		**5**							**5**
Rainbow Smelt		**5**		**5**		**3**			**13**
Steelhead	**12**	**13**	**15**	**15**	**7**	**15**	**14**	**15**	**106**
<400						1			1
>400	12	13	15	15	7	14	14	15	105
Round Goby	**15**	**10**	**14**	**9**	**15**	**2**	**13**		**78**
<60	5		5	5	5	1	5		26
>100	5	5	5		5	1	3		24
60-100	5	5	4	4	5		5		28
Slimy Sculpin		**1**	**5**	**5**					**11**
Spottail Shiner			**1**	**2**			**5**		**8**
Yellow Perch	**5**		**15**	**5**	**29**	**1**	**2**	**13**	**70**
<200				5		1	2		8
>200	5		15		29			13	62
Grand Total	**90**	**131**	**177**	**143**	**112**	**112**	**123**	**99**	**987**

Table 2. Stable Isotope niche overlap. Percentages indicate the probability that an individual of species A (rows) will occur in the 95% isotopic niche area occupied by species B (columns).

	Species B				
Species A	**BRT**	**CHS**	**COS**	**LAT**	**RBT**
BRT		71%	40%	44%	74%
CHS	98%		69%	28%	91%
COS	82%	79%		3%	92%
LAT	61%	17%	2%		18%
RBT	94%	85%	77%	19%	

Table 3. Fatty acid prey classification

	Predicted to be:						
	Alewife	Bloater	Deepwater Sculpin	Rainbow Smelt	Round Goby	Slimy Sculpin	Spottail Shiner
Alewife	21	5	0	0	0	0	0
Bloater	2	10	0	0	0	0	0
Deepwater Sculpin	0	0	15	0	0	0	0
Rainbow Smelt	0	0	1	12	0	0	0
Round Goby	0	0	0	0	77	1	0
Slimy Sculpin	0	0	0	0	0	10	1
Spottail Shiner	0	0	0	0	0	0	8

Table 4. Fatty Acids (means) indicated by SIMPER to describe differences between alewife and round goby. SIMPER provides a % of the difference explained by each fatty acid cumulatively.

	Alewife	Round Goby	Cumulative % of Diff.
20:5n-3	6.2	12.0	12
18:1n-9	14.5	9.6	24
22:6n-3	13.9	8.8	36
16:1n-7	3.5	8.8	47

Table 5. Fatty acid classifications of Chinook Salmon and Lake Trout

		Predicted to be:							
		Early				Late			
		Chinook Salmon							
		NE	NW	SE	SW	NE	NW	SE	SW
Early	NE	0	0	3	0	0	1	0	0
	NW	0	26	1	1	0	0	0	0
	SE	2	1	21	2	0	1	0	0
	SW	0	7	3	15	0	2	0	0
Late	NE	0	0	0	0	17	0	3	7
	NW	0	0	0	0	1	27	1	0
	SE	0	0	0	0	3	3	16	6
	SW	0	1	0	0	4	5	7	3
		Lake Trout							
Early	NE	15	0	0	0	0	0	0	0
	NW	0	10	0	3	0	0	0	0
	SE	0	0	27	2	0	0	0	0
	SW	0	2	2	14	0	0	0	0
Late	NE	0	0	0	0	0	0	4	0
	NW	0	2	0	1	1	11	2	2
	SE	0	0	1	0	1	6	16	5
	SW	0	0	2	0	0	0	5	10

Figures

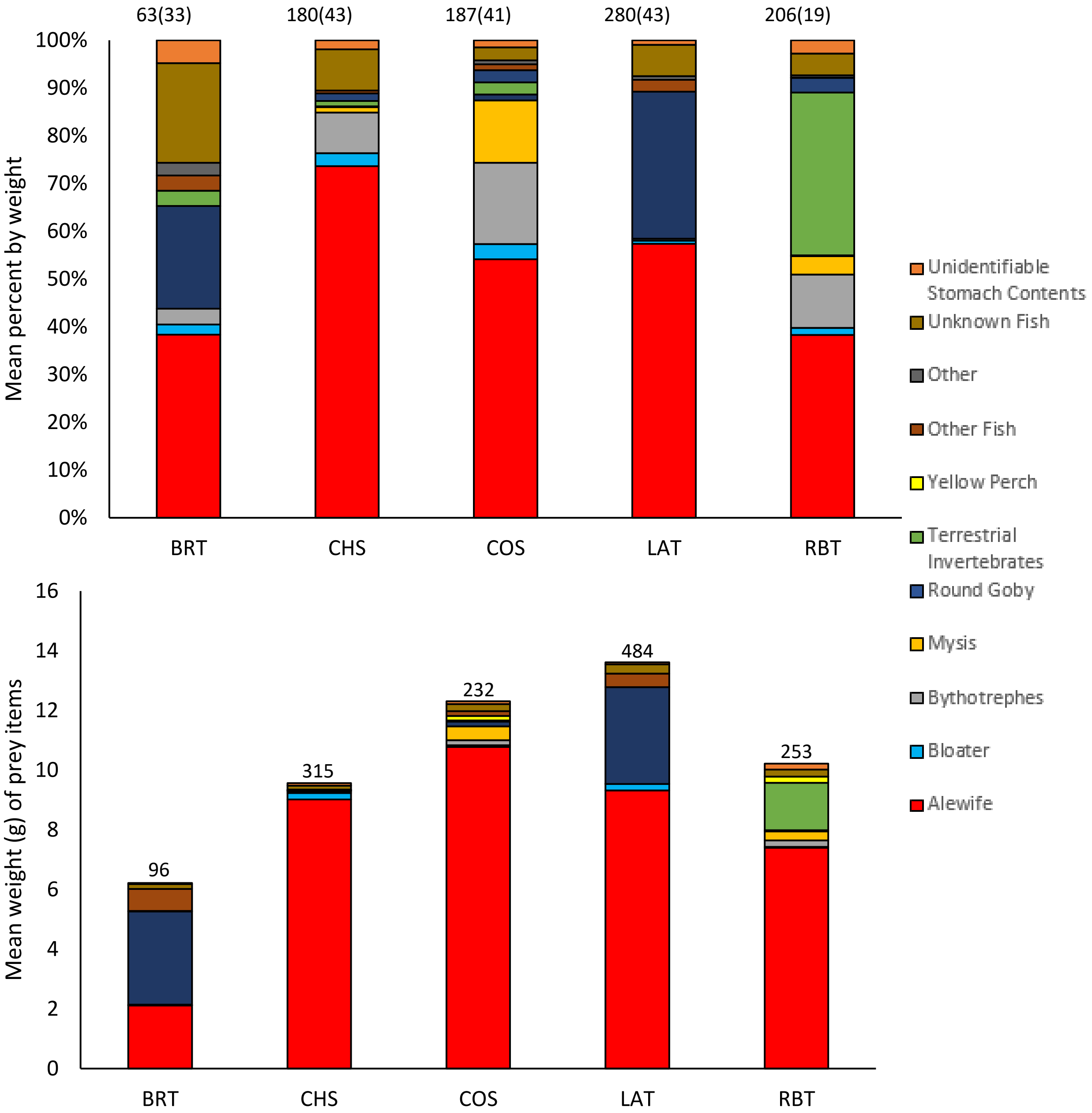

Figure 1. Salmonine Diet Composition

Mean percent diet composition by weight for each salmonine species (top) and mean weight of each prey category for each salmonine species (bottom). Numbers above bars in top figure represent the number of full stomachs and percent that were empty in parentheses and for the bottom figure they represent total number of stomachs analyzed.

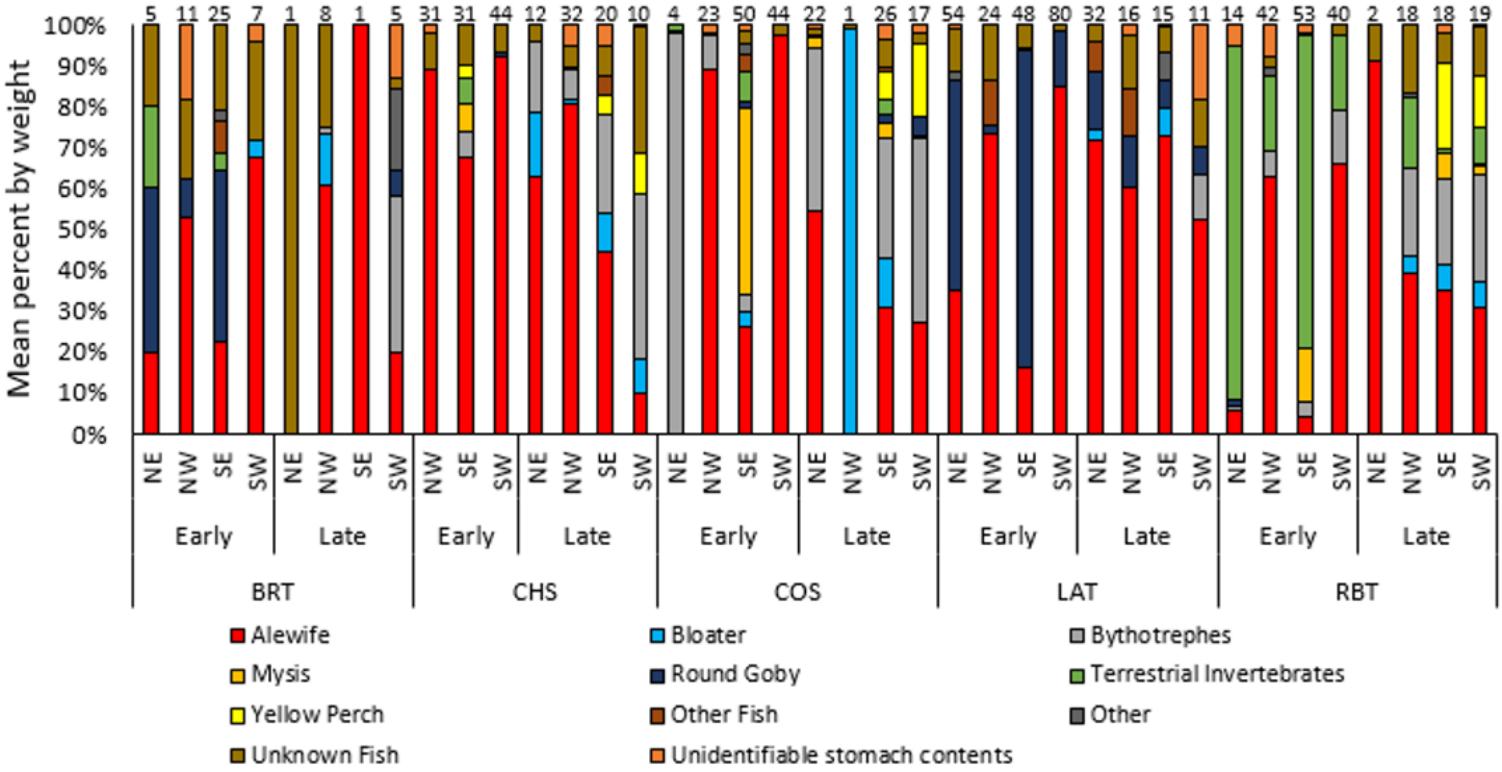

Figure 2. Salmonine Diet Composition

Seasonal and regional patterns of mean percent diet composition by weight for Lake Michigan salmonines. Numbers above bar represent the total number of full stomachs examined.

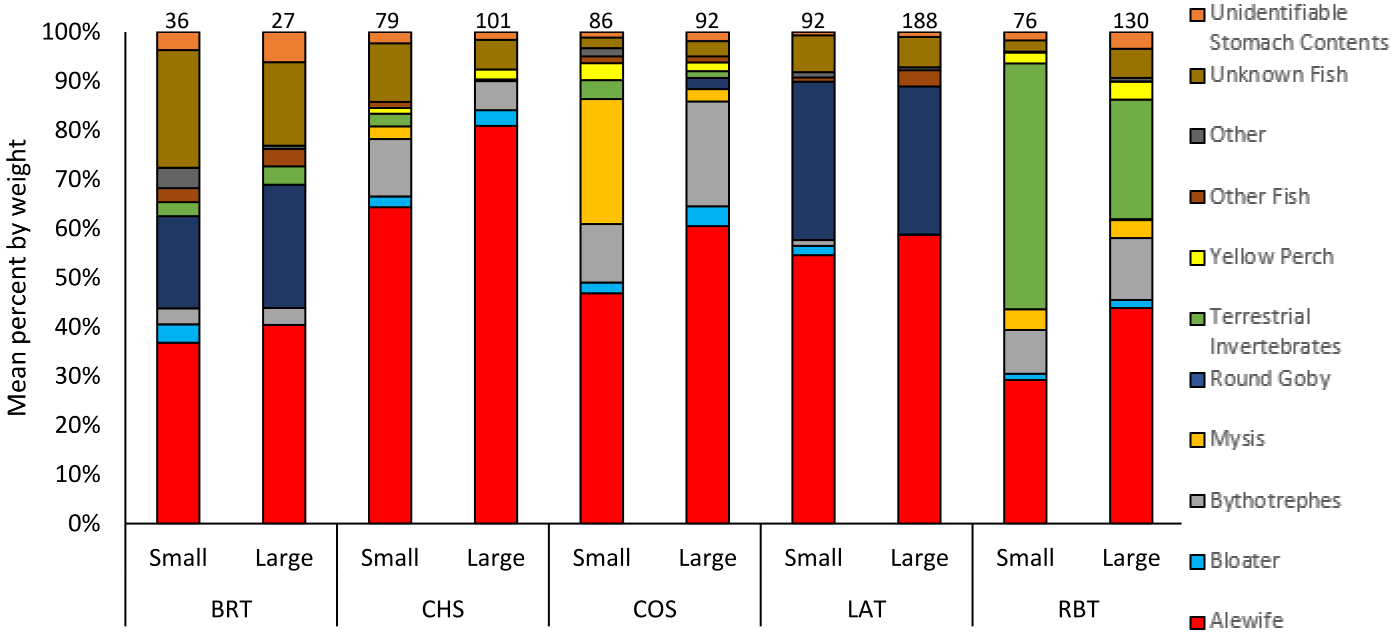

Figure 3. Salmonine Diet Composition

Mean percent diet composition by weight for small (<600 mm) and large (≥600 mm) salmonines. Numbers above bars represent the number of full stomachs analyzed.

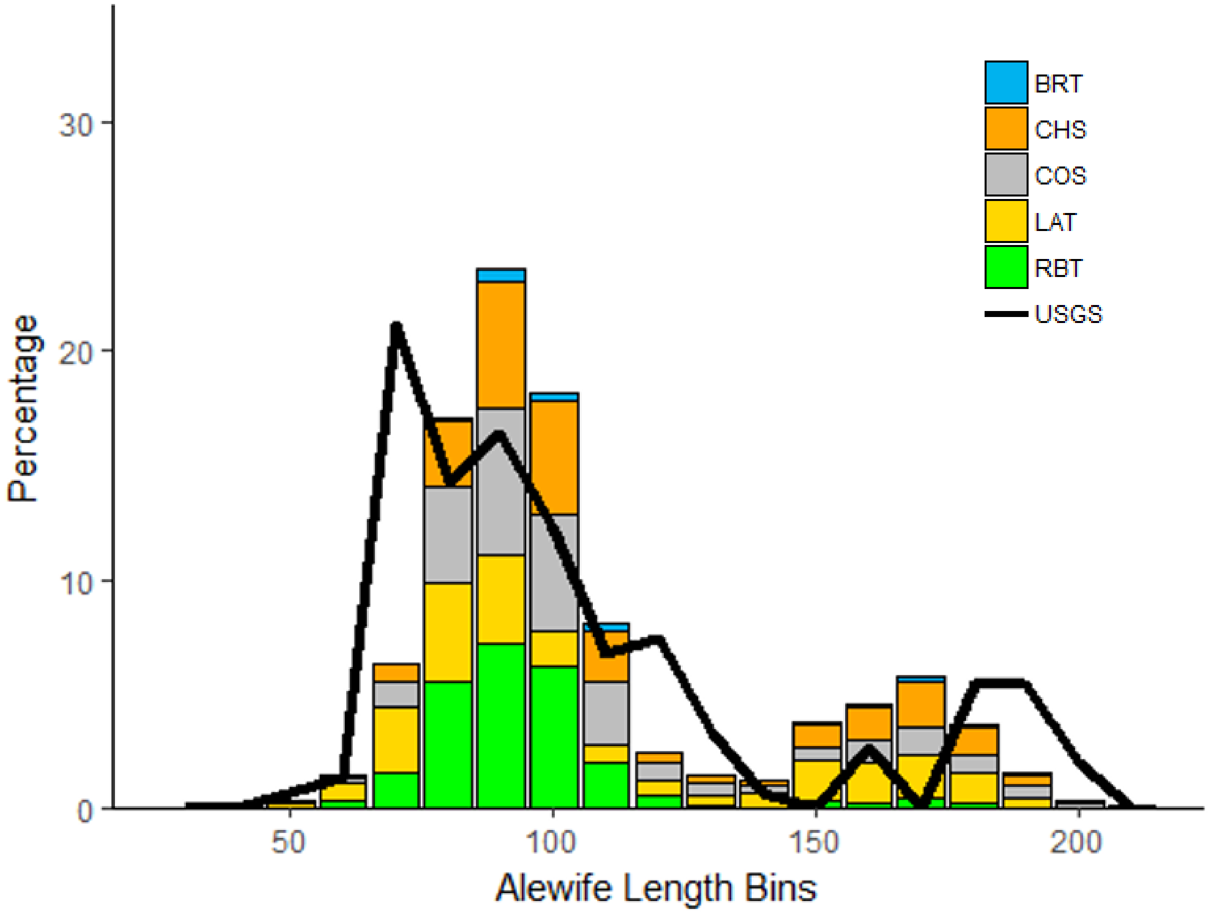

Figure 4. Length Frequency of Alewife in Diets.

Length-frequency distributions (percent of total number) for alewives consumed separated by species. Additionally, length frequencies of alewife collected in annual USGS September trawl surveys in 2016 are included (black line; B. Bunnell, USGS, pers. comm).

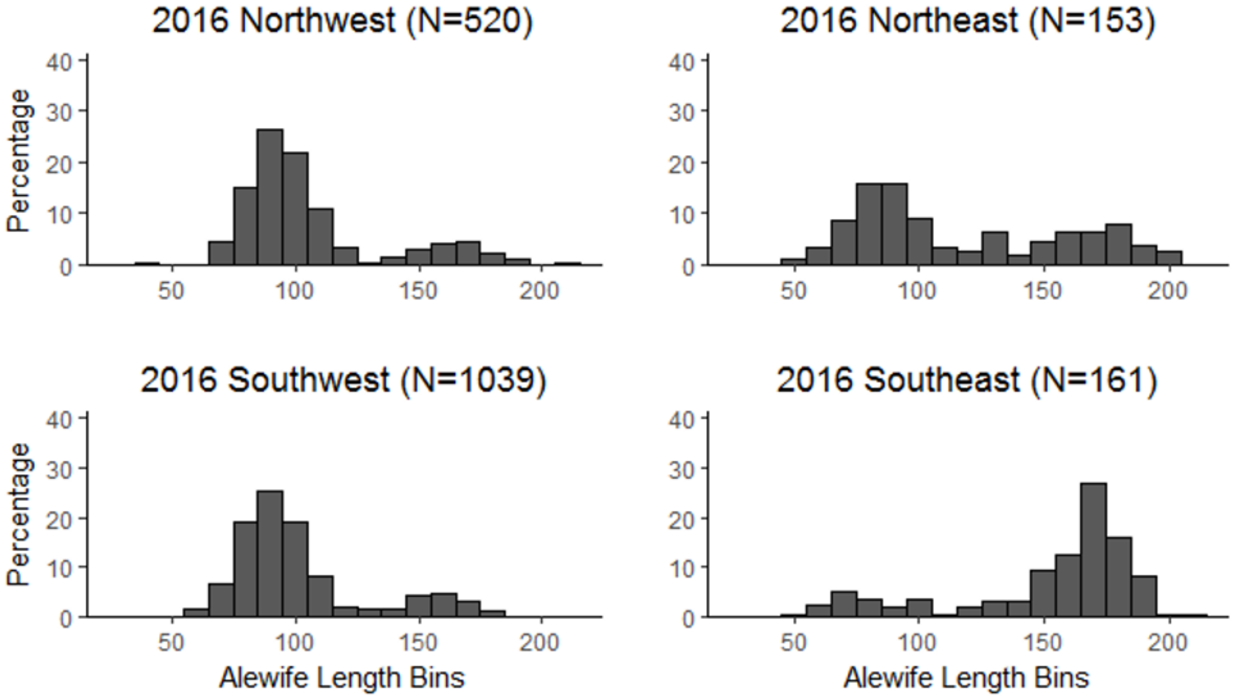

Figure 5. Length Frequency of Alewife in Diets.

Regional length-frequency distributions (percent of total number) for alewives consumed by the five salmonine species. N represents the total number of measurable alewife consumed by salmonines in that region.

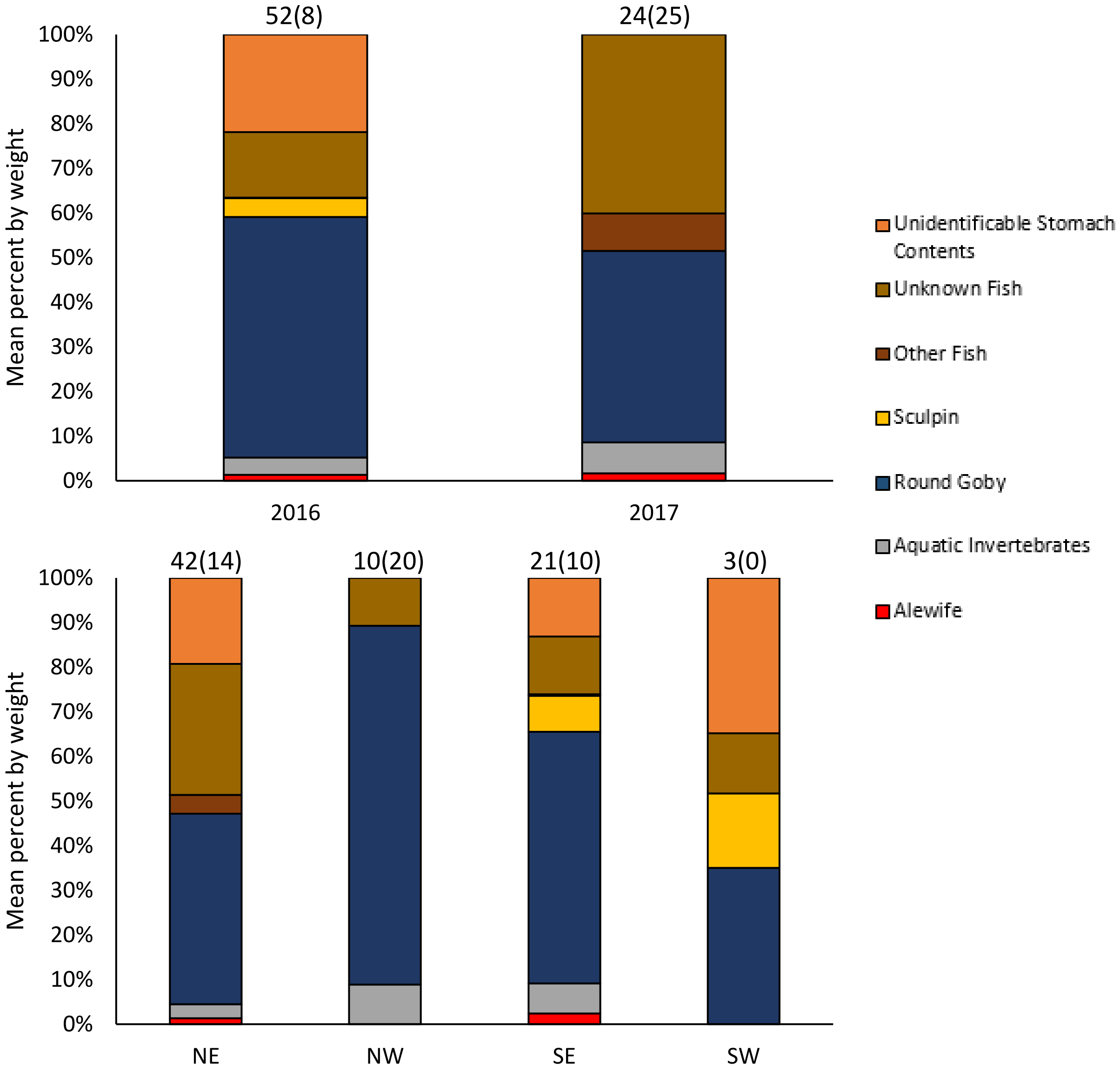

Figure 6. Burbot Diet Composition

Mean lake wide (2016 and 2017 separately; top) and regional (bottom) percent diet composition by weight for burbot. Numbers above bars represent the number of full stomachs and percent that were empty in parentheses.

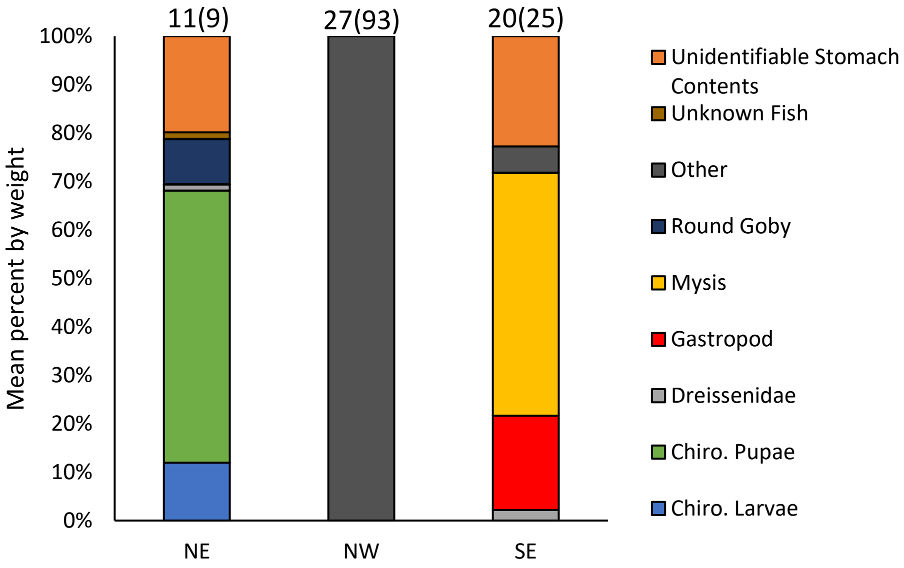

Figure 7. Lake Whitefish Diet Composition

Mean regional percent diet composition by weight for lake whitefish. Numbers above bars represent the number of full stomachs and percent that were empty in parentheses.

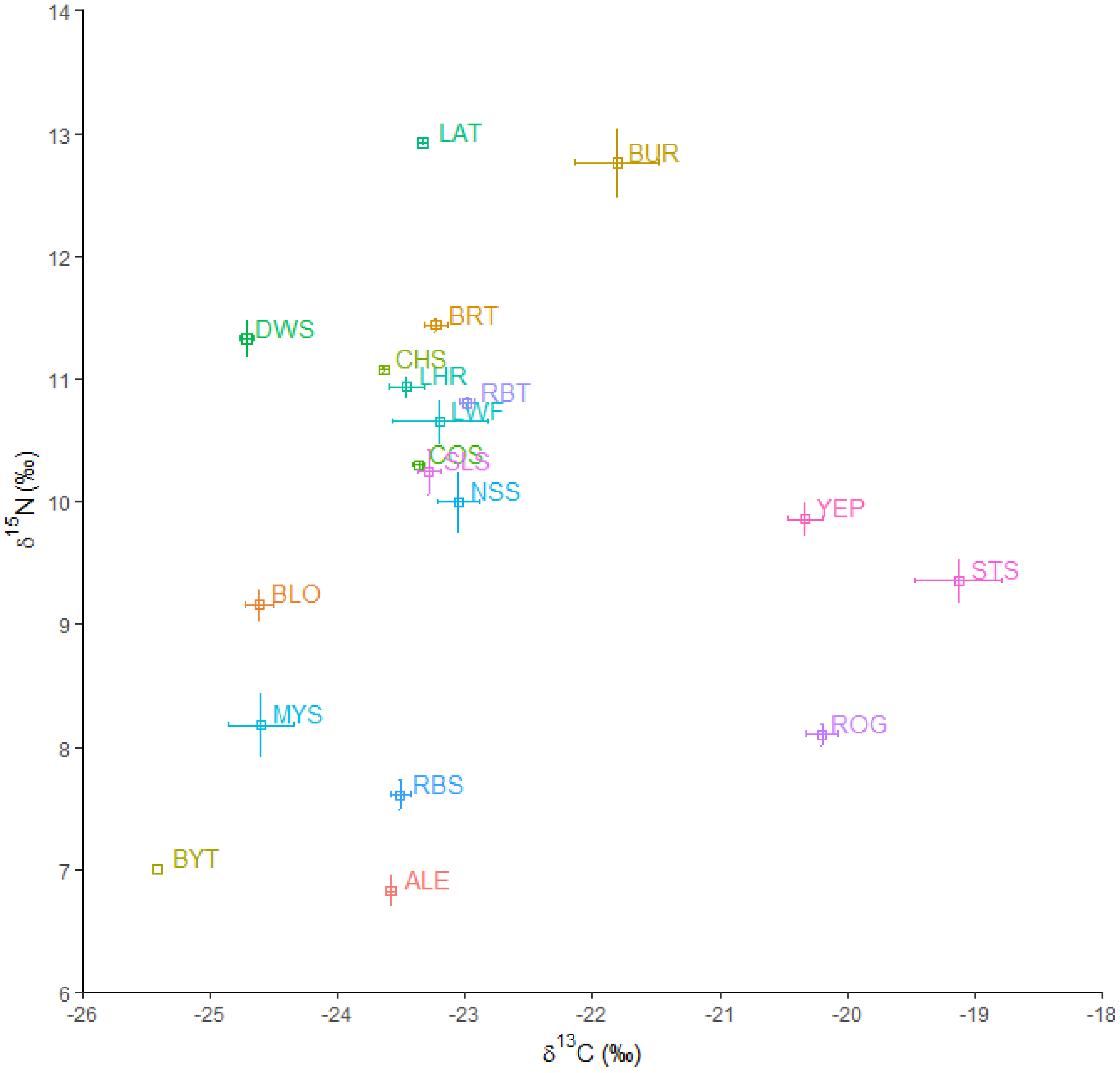

Figure 8. Stable Isotope Biplot.

Mean (± SE) δ^{13}C and δ^{15}N for Lake Michigan fishes and invertebrates. Species include Alewife, ALE; Bloater, BLO; Brown Trout, BRT; Burbot, BUR; *Bythotrephes*, BYT; Chinook Salmon, CHS; Cisco, LHR; Coho Salmon, COS; Deepwater Sculpin, DWS; Lake Trout, LAT; Lake Whitefish, LWF; *Mysis,* MYS; Ninespine Stickleback, NSS; Rainbow Smelt, RBS; Round Goby, ROG; Slimy Sculpin, SLS; Spottail Shiner, STS; Steelhead, RBT; and Yellow Perch, YEP.

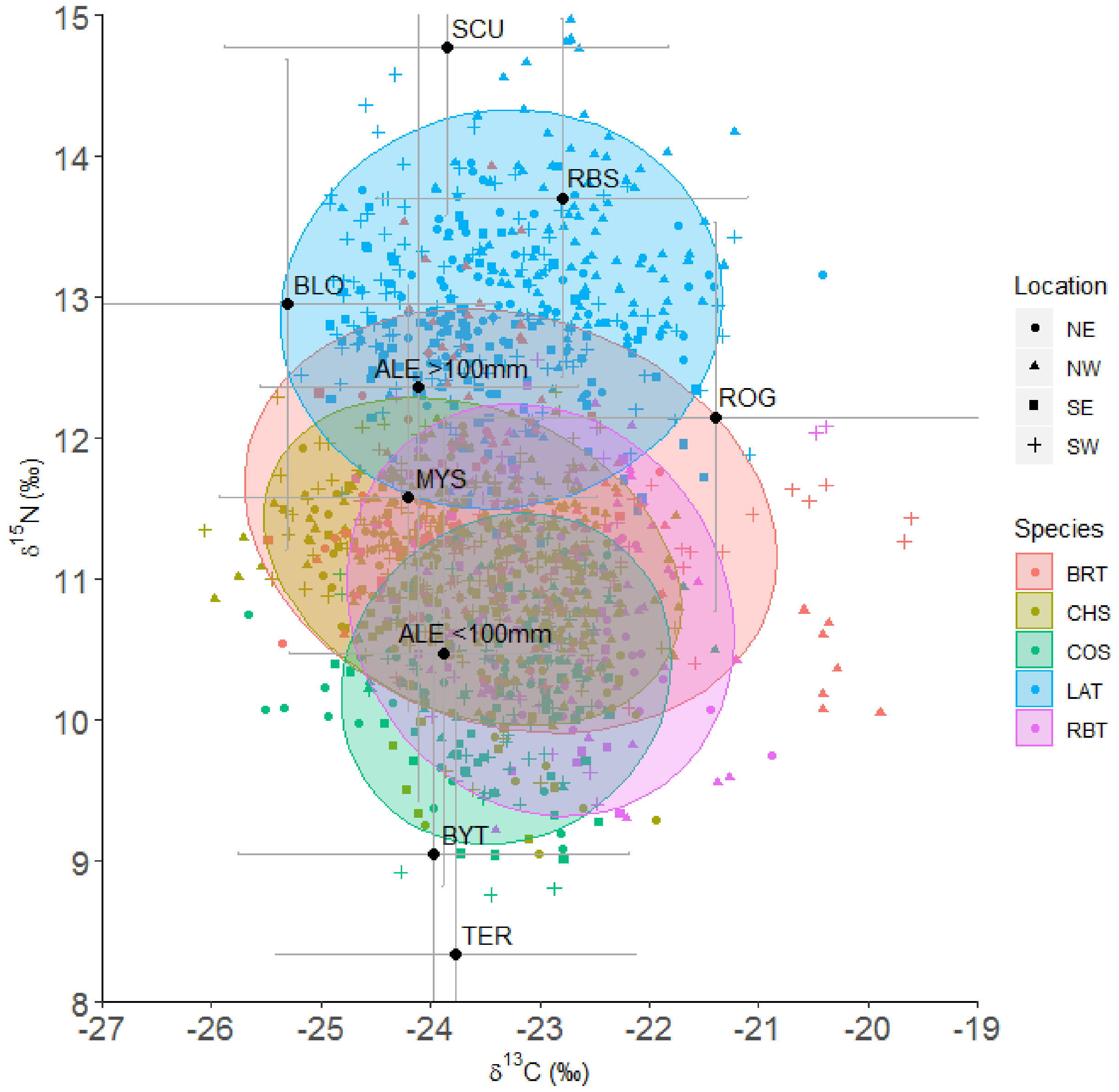

Figure 9. Stable Isotope Niche Overlap

δ^{13}C and δ^{15}N of Lake Michigan Salmonines across study regions. Ellipses encompass 95% of data. Mean (± SD) δ^{13}C and δ^{15}N of prey sources after accounting for trophic enrichment (+0.8‰ and +3.4‰, respectively) were overlaid. Abbreviations are the same as Fig. 7 with the addition of Terrestrial Invertebrates, TER.

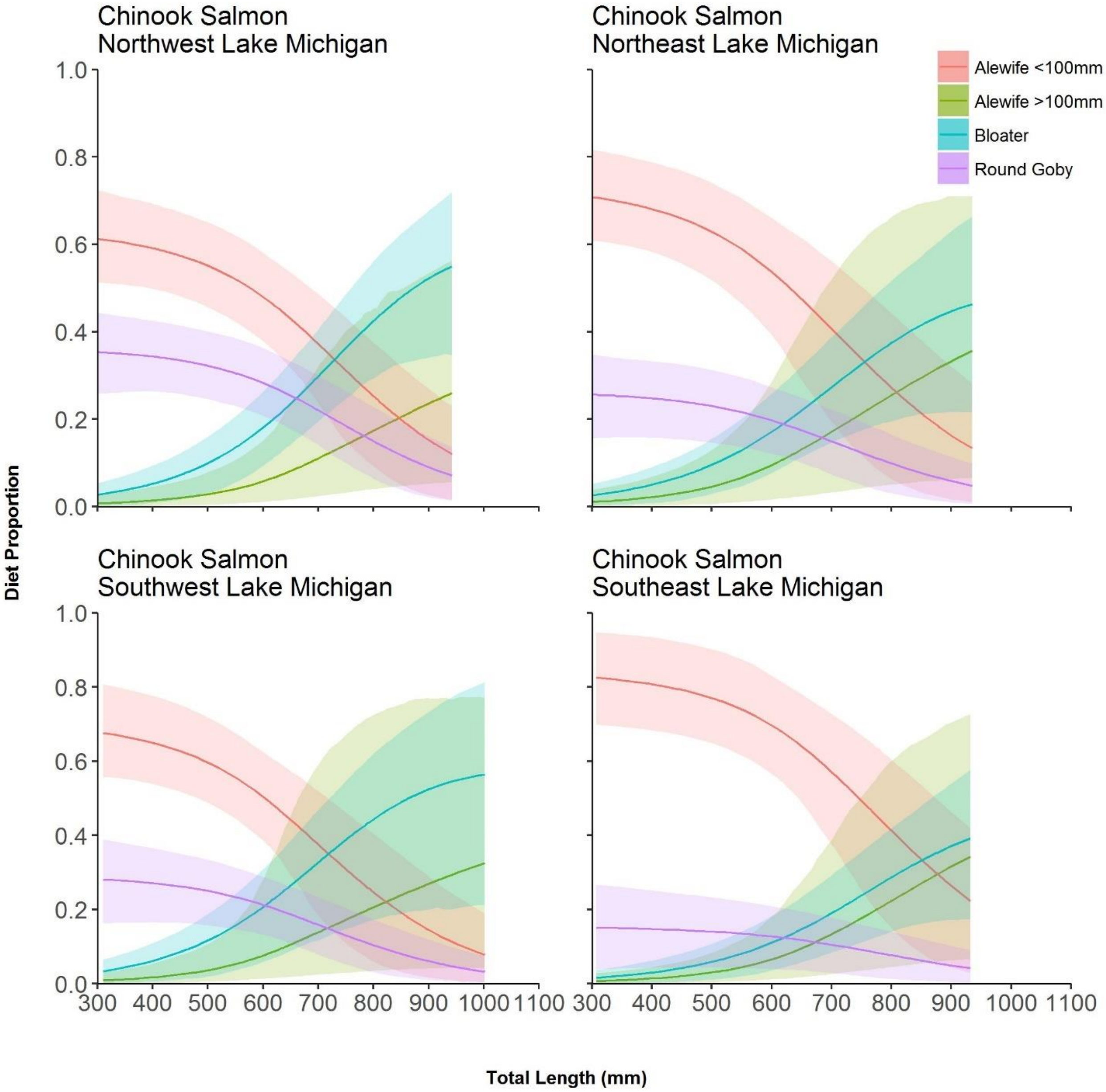

Figure 10. Chinook Salmon SI Mixing Model.

δ^{13}C and δ^{15}N mixing model predictions of dietary proportions as a function of total length (mm) for Chinook Salmon in four study regions of Lake Michigan. Lines represent the mean of the posterior probability distribution and the shaded area represent the bounds of the 95% credible interval of the predicted diet proportion. Line type and colors correspond to specific prey categories.

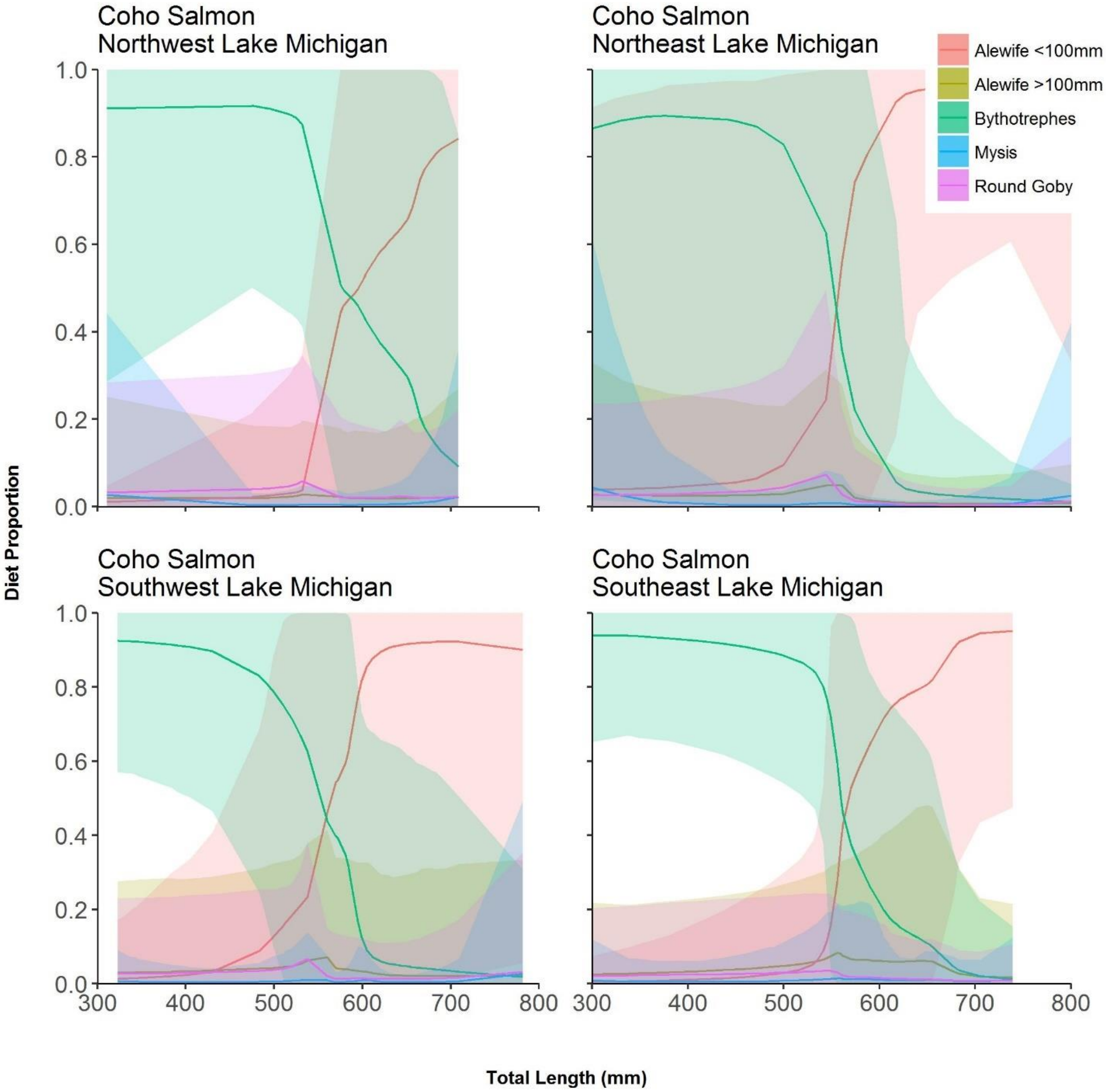

Figure 11. Coho Salmon SI Mixing Model.

δ^{13}C and δ^{15}N mixing model predictions of dietary proportions as a function of total length (mm) for Coho Salmon in four study regions of Lake Michigan. Lines represent the mean of the posterior probability distribution and the shaded area represent the bounds of the 95% credible interval of the predicted diet proportion. Line type and colors correspond to specific prey categories.

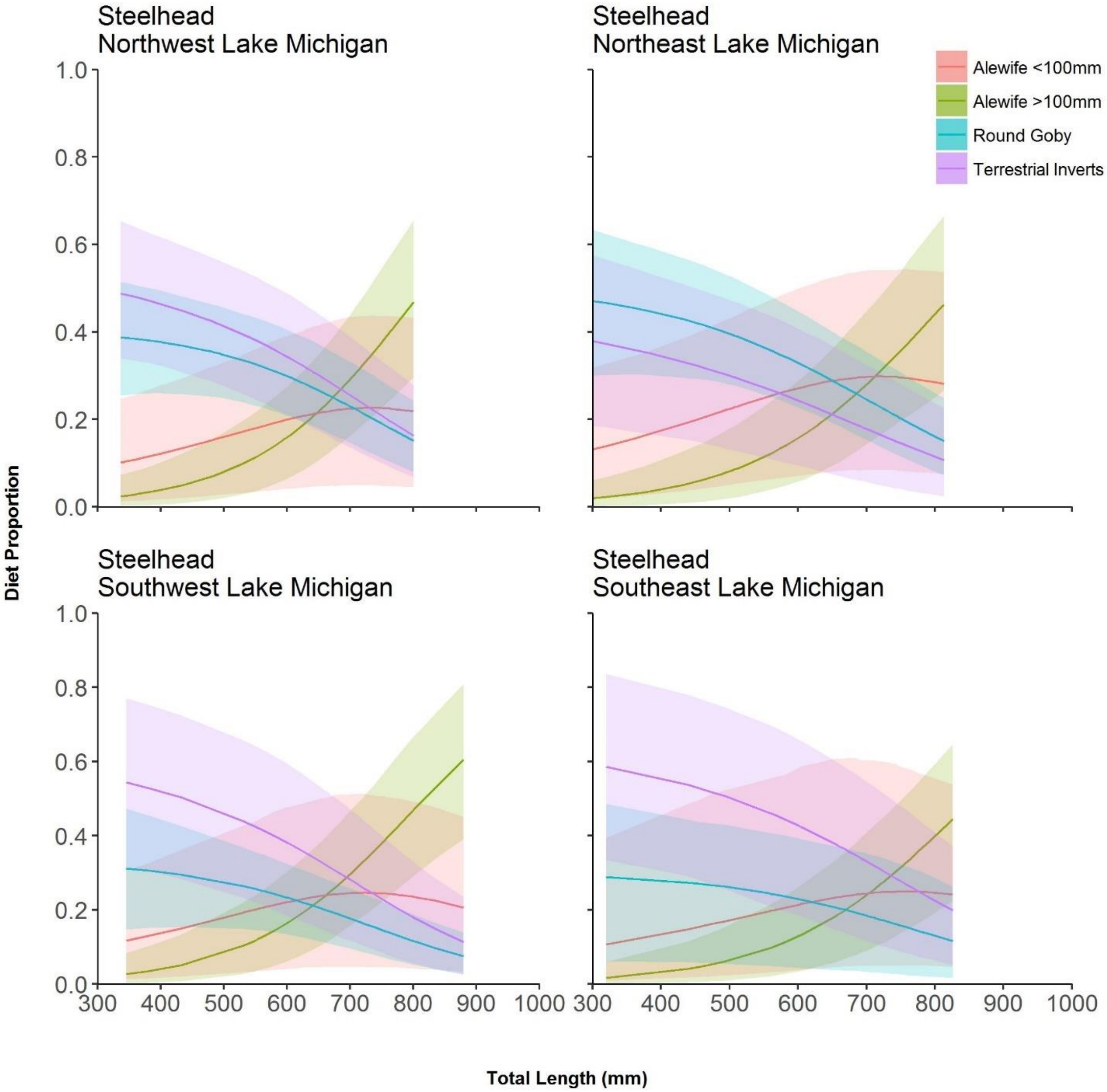

Figure 12. Steelhead SI Mixing Model.

δ^{13}C and δ^{15}N mixing model predictions of dietary proportions as a function of total length (mm) for Steelhead in four study regions of Lake Michigan. Lines represent the mean of the posterior probability distribution and the shaded area represent the bounds of the 95% credible interval of the predicted diet proportion. Line type and colors correspond to specific prey categories.

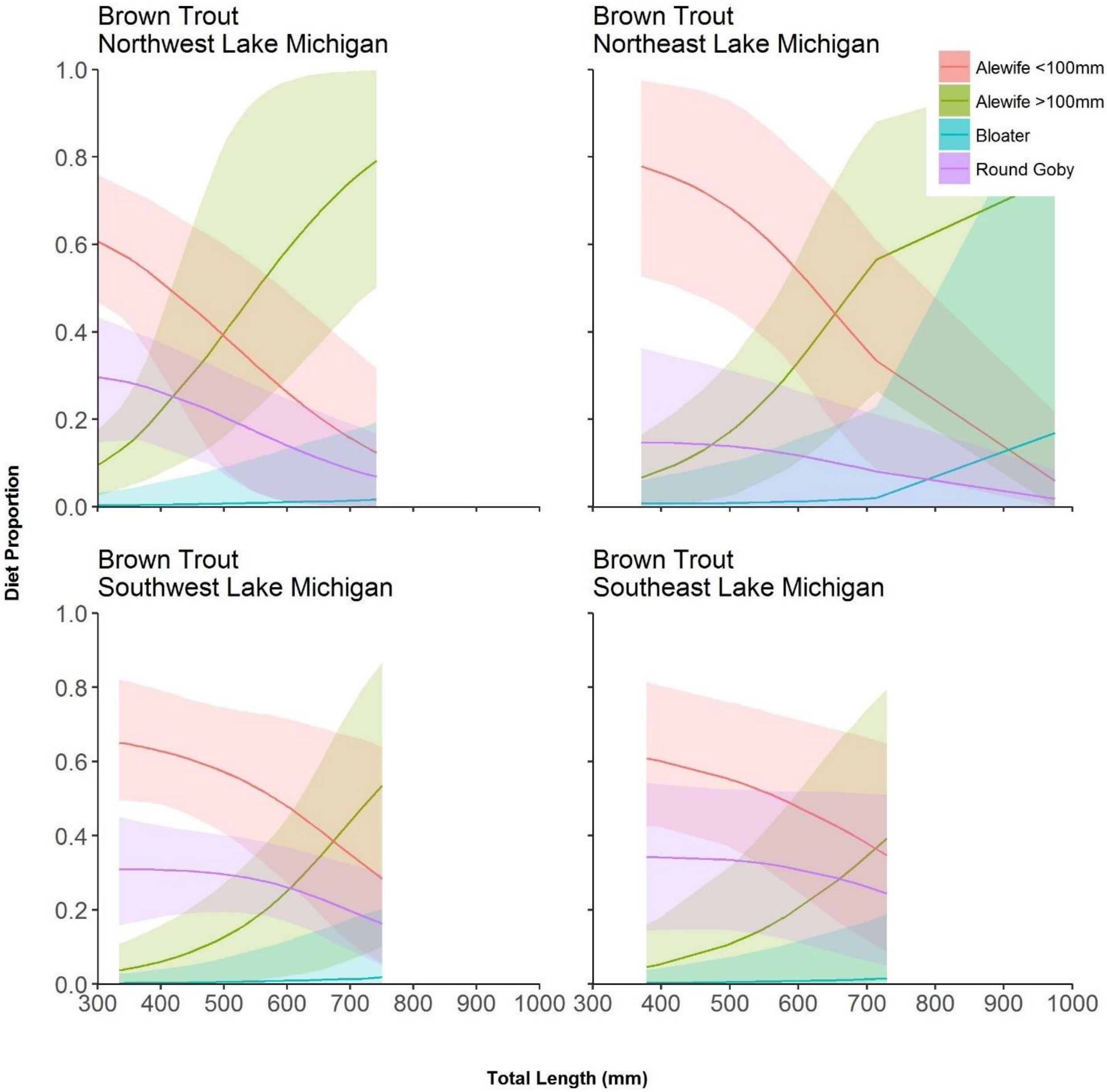

Figure 13. Brown Trout SI Mixing Model.

δ^{13}C and δ^{15}N mixing model predictions of dietary proportions as a function of total length (mm) for Brown Trout in four study regions of Lake Michigan. Lines represent the mean of the posterior probability distribution and the shaded area represent the bounds of the 95% credible interval of the predicted diet proportion. Line type and colors correspond to specific prey categories.

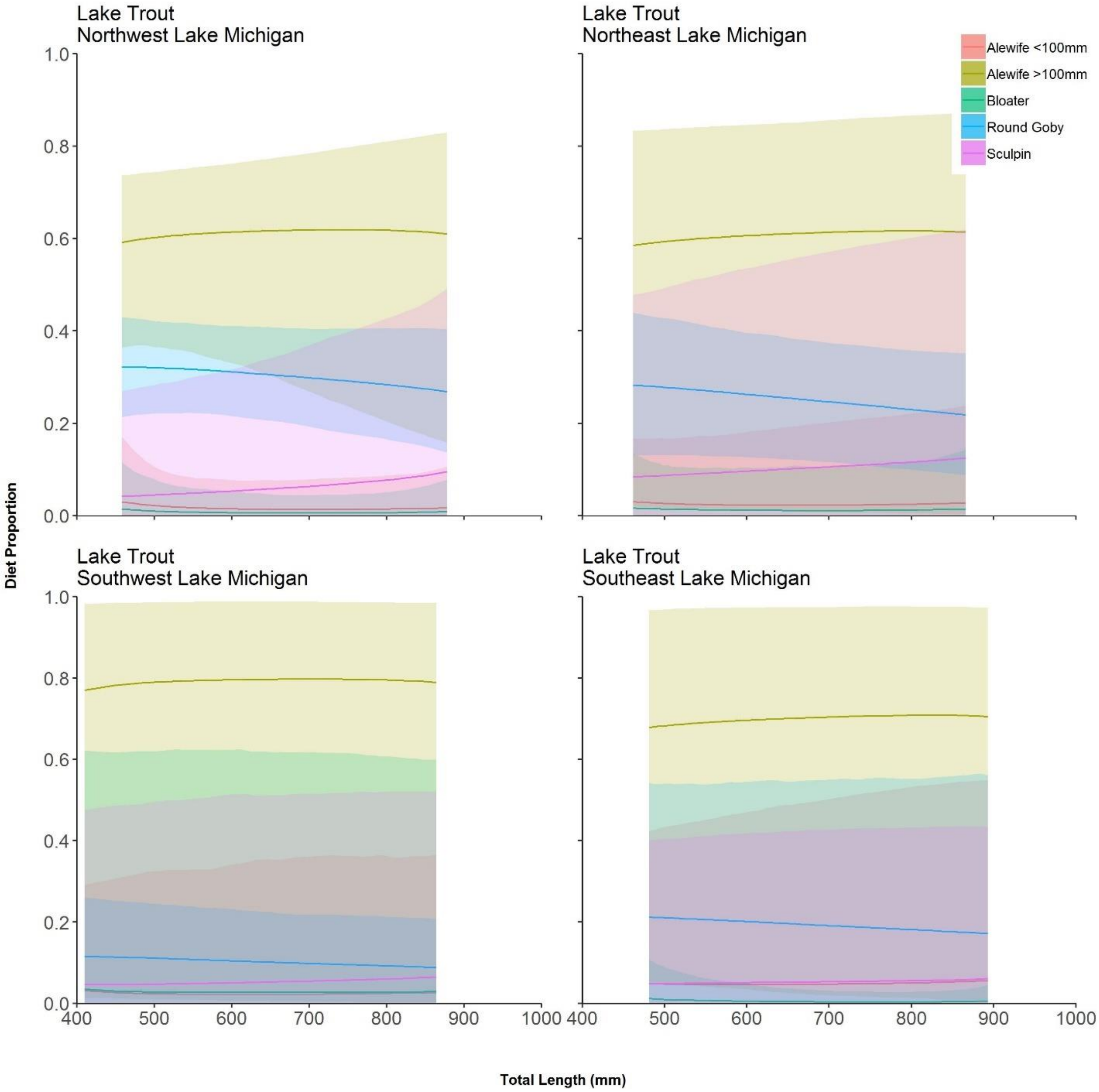

Figure 14. Lake Trout SI Mixing Model.

δ^{13}C and δ^{15}N mixing model predictions of dietary proportions as a function of total length (mm) for Lake Trout in four study regions of Lake Michigan. Lines represent the mean of the posterior probability distribution and the shaded area represent the bounds of the 95% credible interval of the predicted diet proportion. Line type and colors correspond to specific prey categories.

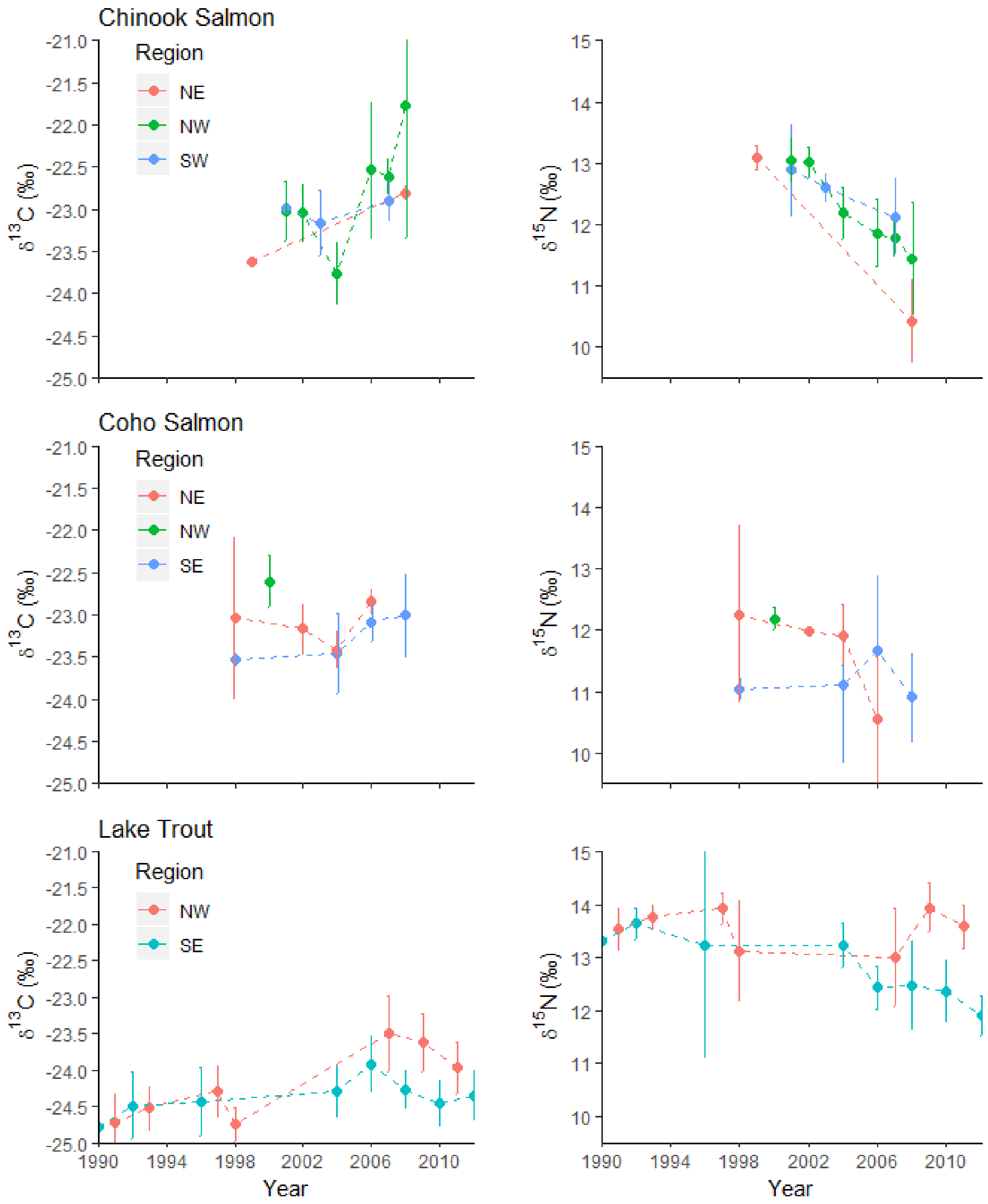

Figure 15. Historic SI Patterns.

Mean (±SD) δ^{13}C (left panels) and δ^{15}N (right panels) of Chinook Salmon (Top), Coho Salmon (Middle), and Lake Trout (bottom) from 1990-2012. Colors correspond to sampling regions.

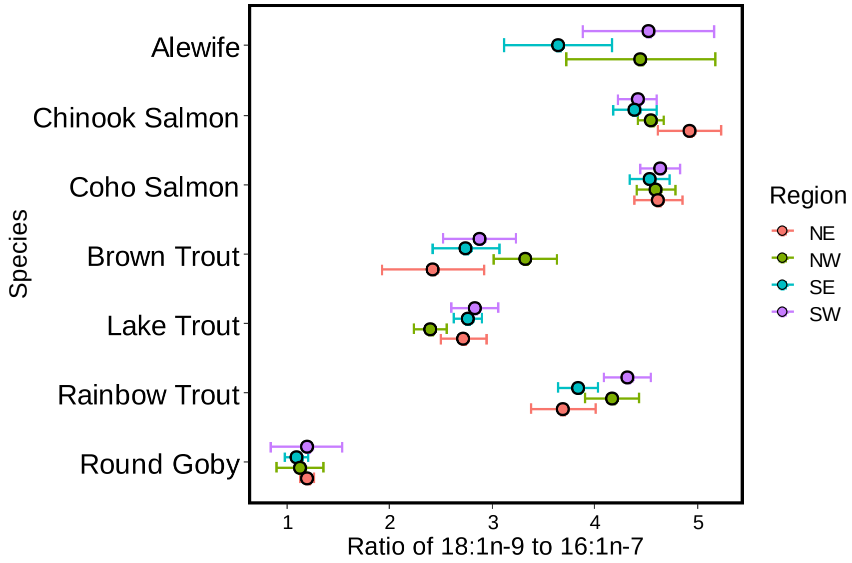

Figure 16. Ratio of 18:1n-9 to 16:1n-7

Plot of the mean and 95% confidence interval of a ratio used to differentiate between the reliance on Alewife and Round Goby as prey items. The ratio of 18:1n-9 to 16:1n-7 has been shown in feeding experiments to represent differences between these two prey items well.

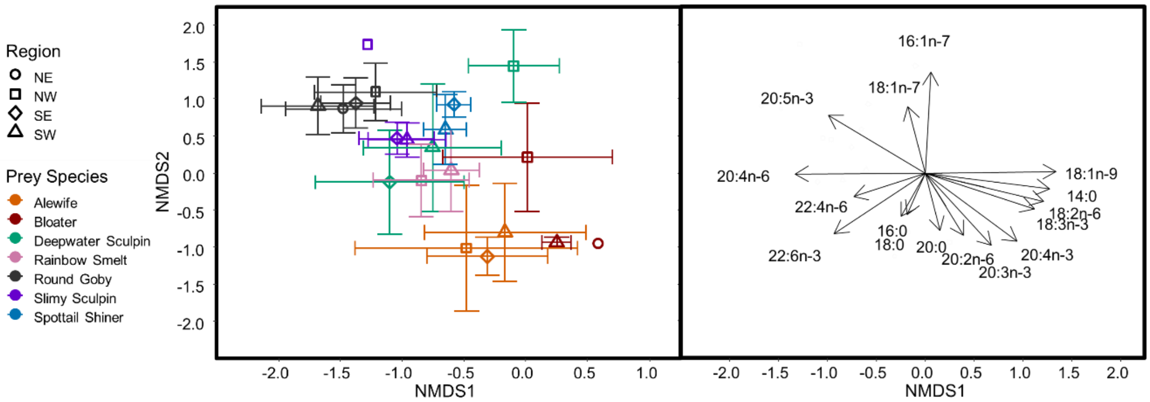

Figure 17. NMDS FA Profiles

NMDS plot of prey specie fatty acid profiles. Colors and symbols used to differentiate between species and regions. Vectors generated using full NMDS data point coordinates and thus can be used here and in Figure 4.

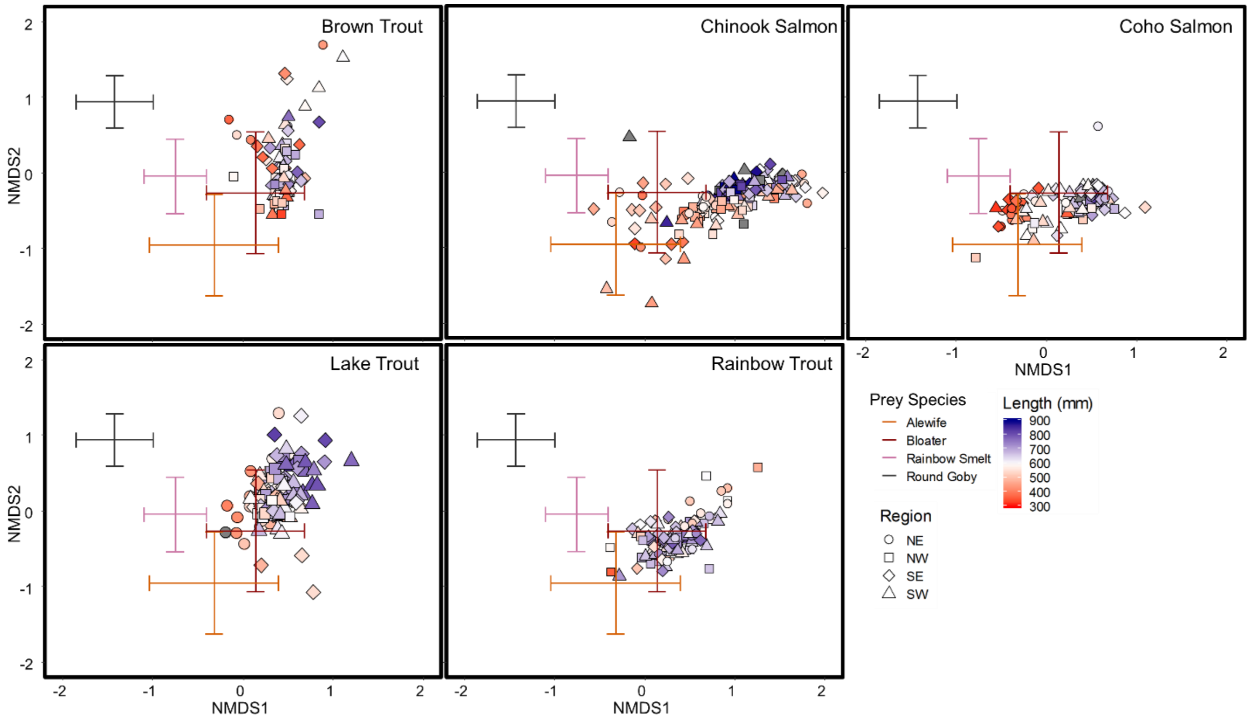

Figure 18. NMDS FA Profiles

NMDS plots of each salmon species showing fatty acid shifts with length and location of the individual. Prey species summaries maintained to provide reference points to figure 3. NMDs points included here were also used in Figure 3, thus vector analysis and figure 3 can be applied here to these figures.

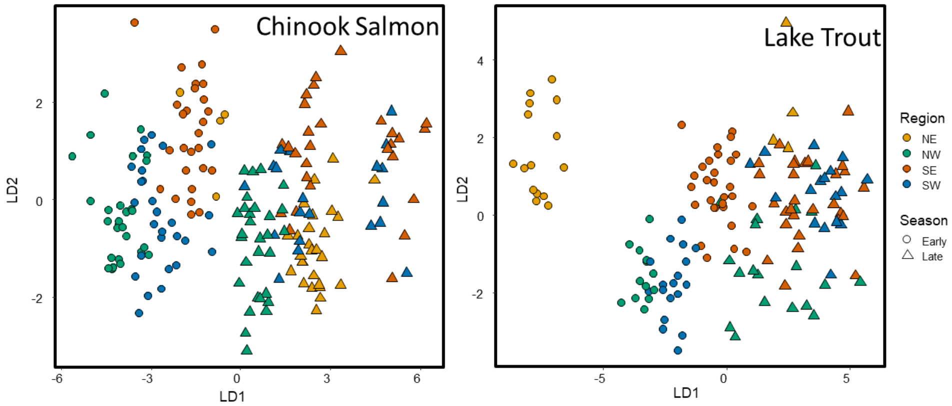

Figure 19. Discriminant Analysis FA Profiles

Visual results of linear discriminant analysis for the combined factors of Season and Region, for Chinook Salmon and Lake Trout. Fatty acid profiles of both species are different by season, and within season show regional differences.