Site C Clean Energy Project

Fisheries and Aquatic Habitat Monitoring and Follow-up Program

Site C Stable Isotope Analysis and Food Web Characterization

Construction Year 6 (2020)

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TECHNICAL MEMORANDUM

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TO Brent Mossop and Nich Burnett; BC Hydro

CC Dustin Ford

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BC HYDRO SITE C STABLE ISOTOPE ANALYSIS AND FOOD WEB CHARACTERIZATION

BC Hydro requested Golder Associates Ltd. (Golder) summarize stable isotope data collected during the Site C Clean Energy Project's (the Project) Peace River Large River Fish Indexing Survey (Mon-2, Task 2a of the Site C Fisheries and Aquatic Habitat Monitoring and Follow-up Program [FAHMFP]) and the survey's predecessor under the Peace River Water Use Plan, the Peace River Fish Index. This summary is intended to help address questions that BC Hydro has received regarding the existing food web structure in Dinosaur Reservoir and the Peace River and is intended to provide summarized background data that could be used in the future to identify potential changes to food web structure in the Peace River in response to the construction or operation of the Project.

1.0 BACKGROUND

The Site C Clean Energy Project, including Project construction, reservoir filling, and operation, could affect fish and fish habitat via three key pathways: changes to fish habitat (including nutrient concentrations and lower trophic biota), changes to fish health and fish survival, and changes to fish movement. These pathways are examined in detail in Volume 2 of the Project's Environmental Impact Statement (EIS; BC Hydro 2013). The EIS makes both qualitative and quantitative predictions of fish production in the Peace River downstream of the Project.

Quantitative predictions of fish biomass downstream of the Project were generated as part of the EIS. For these predictions, each fish species was assigned to one of four groups:

- Group 1 consisted of large-bodied fish typically targeted by anglers (i.e., Burbot [*Lota lota*], Goldeye [*Hiodon alosoides*], Lake Trout [*Salvelinus namaycush*], Northern Pike [*Esox lucius*], Rainbow Trout [*Oncorhynchus mykiss*], and Walleye [*Sander vitreus*])
- Group 2 included species considered "passage sensitive" (i.e., Arctic Grayling [*Thymallus arcticus*], Bull Trout [*Salvelinus confluentus*], and Mountain Whitefish [*Prosopium williamsoni*])
- Group 3 included planktivorous species (i.e., Kokanee [*Oncorhynchus nerka*] and Lake Whitefish [*Coregonus clupeaformis*])
- Group 4 fish consisted of all remaining species (i.e., Northern Pikeminnow [*Ptychocheilus oregonensis*], sucker species, and small-bodied fish species)

Relative to pre-Project estimates, the EIS predicts decreased biomass of Group 1 fishes over the short- (10 years) and long-term (greater than 30 years), increased biomass of Group 2 fishes over the short- and long-term, similar biomasses of Group 3 fishes over the short- and long-term, and decreased biomass of Group 4 fishes over the short- and long-term. There is uncertainty around how changes to biomass of the different groups of fish may affect food web structure; however, changes to the abundance of different consumer groups could result in changes in prey abundance and consumer feeding patterns.

1.1 Objective

The objective of this memo is to analyze existing literature, stable isotope data, mercury data, and stomach content data to generate a baseline food web in Dinosaur Reservoir and the Peace River (data collected from 2010-2019). There are no hypotheses within the FAHMFP that specifically relate to the Peace River food web's response to the construction or operation of the Project; however, food web data may be important to answering the FAHMFP's Peace River Fish Community Monitoring Program's (Mon-2) overarching management question:

1) How does the Project affect fish in the Peace River between the Project and the Many Islands area in Alberta during the short (10 years after Project operations begin) and longer (30 years after Project operations begin) term?

2.0 OVERVIEW OF ANALYSIS

2.1 Stable Isotope Analysis

Analysis of naturally occurring stable isotopes in fish and their potential prey can provide information on feeding linkages. Fractionation of the heavy and rare nitrogen isotope (^{15}N) compared to the light and abundant nitrogen isotope (^{14}N) can be used to infer trophic level (e.g., who is eating what type(s) of prey), and fractionation of the heavy and rare carbon isotope (^{13}C) compared to the light and abundant carbon isotope (^{12}C) can be used to inform carbon source or location of prey sources (e.g., who is eating where). Stable isotope composition is presented as the ratio of the rare to abundant isotope normalized to a known reference, and is expressed in parts per thousand, or "per mil" units (denoted ‰). Isotopic ratios calculated in this way (also sometimes referred to as a "signature") are denoted $\delta^{15}N$ (read "delta N-15") and $\delta^{13}C$ (read "delta C-13").

Isotopic signatures in muscle tissue reflect the diet assimilated over at least several months in small, fast growing fish and longer periods in larger, slow growing fish (Vander Zanden et al. 2015). As a result, stable isotope analysis provides a time-integrated measure of an organism's trophic position and accounts for temporal and spatial variability in feeding dynamics (Post 2002). This time integration provides an advantage compared to stomach content analysis, which reflects a shorter-term diet and may not reflect variation in digestibility and assimilation of source items (Perkins et al. 2014).

The relative abundance of the heavy nitrogen isotope increases with trophic level, which allows the use of $\delta^{15}N$ as a measure of trophic position (Cabana and Rasmussen 1996; Post et al. 2002). Comparisons between food webs in different ecosystems can be conducted by considering the $\delta^{15}N$ signature of nitrogen at the base of the food web (Cabana and Rasmussen 1996). The relative enrichment of $\delta^{15}N$ between trophic levels has been observed to vary between species and ecosystems, but typically enrichment of 2.5‰ to 4‰ is observed between diet and consumer (Post 2002, Perkins et al. 2014). Comparison of $\delta^{15}N$ between species can be used to calculate an organism's trophic level, the overall food chain length, and, in combination with $\delta^{13}C$, can be used to map prey-consumer connections.

In contrast to the stable nitrogen isotope, enrichment of $\delta^{13}C$ from diet to consumer is usually not observed. Rather, the $\delta^{13}C$ signature in consumers directly reflects the carbon signature of their food sources (mean change across trophic levels 0‰ to 0.4‰; Post 2002, Perkins et al. 2014). Previous studies have found that macrophytes collected from the littoral zones of lakes and benthic algae collected from lakes are enriched in ^{13}C compared to the same species in fast flowing rivers (France 1995; Peterson and Fry 1987). Within freshwater lakes, France (1995) found ^{13}C to be enriched in benthic algae and littoral consumers when compared with phytoplankton and pelagic consumers. One hypothesis to explain these occurrences of ^{13}C enrichment is that the lower water turbulence in near-bottom habitats results in a boundary layer around algal cells within which CO_2 becomes depleted and is more slowly replenished than in pelagic habitats, which results in enrichment of ^{13}C in benthic algae due to CO_2 limitation (France 1995). Comparisons of $\delta^{13}C$ between species can be used to infer where consumers are eating prey (e.g., rivers vs. lakes, pelagic vs. benthic or littoral areas). Used in combination with $\delta^{15}N$, $\delta^{13}C$ can be used to map prey-consumer connections.

2.2 Mercury

Similar to $\delta^{15}N$, mercury bioaccumulates in the food chain and is expected to increase with increasing trophic levels. Mercury was used herein as a third environmental tracer, and the accumulation of mercury was compared to $\delta^{15}N$ to understand if differences in bioaccumulation of $\delta^{15}N$ and mercury occurred in any species or locations.

2.3 Stomach Contents

Analysis of the composition of fish stomach contents can provide useful information to evaluate short-term feeding habits. Direct analysis of stomach contents can provide information on the frequency of occurrence, relative composition, and relative importance of different prey items at the time of sampling. Analysis of fish stomach contents provides a "snapshot" of recently consumed prey (Cresson et al. 2014). The benefit of stomach content analysis is the identification of the specific taxa consumed; however, different rates of prey digestion and understanding seasonal and temporal variability in prey consumption can make the interpretation of data difficult. Typically, large numbers of samples are required to obtain a representative view of dietary patterns for a fish population. Stomach content data can be interpreted together with stable isotope data to provide a holistic understanding of dietary preferences (Davis et al. 2012).

3.0 METHODS

3.1 Available Data

Data used for this report were queried from the "BC Hydro Peace River Mercury and SIA Database version 2020.10.20" (the database; Golder 2020). The database includes data collected as part of BC Hydro's Large River Fish Indexing Program (2002 to 2017; P&E and Gazey 2003; Mainstream and Gazey 2004, 2005, 2006, 2007, 2008), the Peace Project Water Use Plan (2008 to 2014; Mainstream and Gazey 2009, 2010, 2011, 2012, 2013, 2014; Golder and Gazey 2015), and the Peace River Large Fish Indexing Survey (2015 to 2019; Golder and Gazey 2016, 2017, 2018, 2019; Golder and Gazey in prep.), collectively referred to as the Indexing Survey, as well as data collected as part of the Site C Reservoir and Peace River Fish Food Organisms Monitoring Program (Mon-6 and Mon-7; Ecoscape 2018). In addition to the database query, fish stomach content data were compiled from the Mon-6 and Mon-7 sampling program (Ecoscape 2018). The below summary is based on fish captured between mid-August and early October in the following portions of the Peace River mainstem:

- Section 1 – an approximately 8 km section downstream of Peace Canyon Dam and adjacent to the community of Hudson's Hope
- Section 3 – an approximately 20 km section immediately downstream of the Halfway River's confluence with the Peace River
- Section 5 – an approximately 14 km section immediately downstream of the Moberly River's confluence with the Peace River
- Section 6 – an approximately 11 km section immediately downstream of the Pine River's confluence with the Peace River
- Section 7 – an approximately 15 km section between the Beatton and Kiskatinaw rivers' confluences with the Peace River
- Section 9 – an approximately 18 km section near the Many Islands area in Alberta

Sections 1 and 3 were sampled during all study years; Section 5 was not sampled in 2003, 2004, or 2006, and Sections 6, 7, and 9 were not sampled prior to 2015. For Sections 6, 7, and 9, the 2009, 2010, and 2011 analyses were supplemented with data from other BC Hydro datasets when possible (i.e., Mainstream 2010, 2011, 2013). All fish were captured using a boat electroshocker using methods detailed in Golder and Gazey (2019). Stable isotope data were only available for 2010, 2011, 2017, 2018, and 2019. Table 1 summarizes the data compiled and reported herein, including fish and invertebrate tissue data queried from the database (Golder 2020) and stomach content data collected as part of Mon-6 and Mon-7 (Ecoscape 2018).

Table 1: Summary of Available Fish and Invertebrate Data

Year	Sample Type	Location	Number of Samples				Data Source (s)
			SIA	Hg	Paired SIA - Hg	Stomach Contents[1]	
2010	Fish	Dinosaur Reservoir	50	49	49	46	Mainstream and Gazey 2011; Ecoscape 2018
		Section 1	0	0	0	4	
		Section 2	0	0	0	36	
		Section 4	0	0	0	16	
		Section 3	43	42	42	0	
		Section 5	11	11	11	0	

Year	Sample Type	Location	Number of Samples				Data Source (s)
			SIA	Hg	Paired SIA - Hg	Stomach Contents[1]	
2011	Fish	Dinosaur Reservoir	2	44	2	50	Mainstream and Gazey 2012; Ecoscape 2018
		Section 1	0	31	0	50	
		Section 2	0	0	0	19	
		Section 3	0	26	0	0	
		Section 5	0	0	0	11	
		Section 7	0	31	0	8	
		Section 8	0	17	0	0	
2017	Fish	Section 1	31	9	8	35	Golder and Gazey 2018; Ecoscape 2018
		Section 2	0	0	0	45	
		Section 3	78	20	20	0	
		Section 4	0	0	0	18	
		Section 5	51	8	8	31	
		Section 6	51	5	5	16	
		Section 7	40	8	8	8	
		Section 9	110	36	36	0	
	Invertebrate	Dinosaur Reservoir	0	2	0	-	Ecoscape 2018
		Section 1	0	1	0	-	
		Section 2	0	3	0	-	
		Section 3	0	1	0	-	
		Section 4	0	2	0	-	
		Section 5	0	4	0	-	
		Section 6	0	1	0	-	
		Section 7	0	1	0	-	
		Section 8	0	2	0	-	
		Section 9	0	2	0	-	

Year	Sample Type	Location	Number of Samples				Data Source (s)
			SIA	Hg	Paired SIA - Hg	Stomach Contents[1]	
2018	Fish	Section 1	43	43	43	28	Golder and Gazey 2019; Ecoscape 2018
		Section 2	0	0	0	48	
		Section 3	76	77	76	0	
		Section 4	0	0	0	16	
		Section 5	77	76	76	23	
		Section 6	37	37	36	4	
		Section 7	28	29	28	6	
		Section 9	103	102	102	0	
	Invertebrate	Dinosaur Reservoir	0	2	0	-	Ecoscape 2018
		Section 1	0	1	0	-	
		Section 2	0	3	0	-	
		Section 4	0	1	0	-	
		Section 5	0	6	0	-	
		Section 6	0	2	0	-	
		Section 7	0	2	0	-	
		Section 8	0	1	0	-	
		Section 9	0	2	0	-	
2019	Fish	Section 1	18	18	18	0	Golder and Gazey 2020
		Section 3	23	23	23	0	
		Section 5	33	33	33	0	
		Section 6	57	57	57	0	
		Section 7	33	33	33	0	
		Section 9	25	25	25	0	

1. Data were queried from BC Hydro Peace River Mercury and SIA Database version 2020.10.20 (Golder 2020), except for stomach content data which was compiled from Ecoscape (2018).

3.2 Data Analysis

3.2.1 Stable Isotope Analysis

Nitrogen

The equation used to derive $\delta^{15}N$ is provided below. For example, a sample with $\delta^{15}N$ of 20‰ has 2% more ^{15}N than the reference material (which is atmospheric N_2), and therefore is described as relatively *enriched* in ^{15}N. Conversely, a sample with $\delta^{15}N$ of -20‰ has 2% less ^{15}N that the reference material, and therefore is described as relatively *depleted* in ^{15}N (Clark 2015).

$$\delta^{15}\mathrm{N} = \left(\frac{(N^{15}/N^{14})_{sample} - (N^{15}/N^{14})_{reference}}{(N^{15}/N^{14})_{reference}}\right) \times 1000‰ \qquad \text{(Equation 1)}$$

$\delta^{15}N$ ranges of prey taxa were predicted for each fish species based on an assumed enrichment of 2.5‰ to 4‰ for each trophic level (Post 2002; Perkins et al. 2015). Predicted prey $\delta^{15}N$ ranges were calculated and compared to the measured $\delta^{15}N$ ranges of each species. Where overlap was observed, the prey taxon was identified as a potential dietary item. A *potential* prey item does not necessarily indicate a biologically *likely* prey item. Biologically unlikely results were identified in the integrated analysis (Section 3.2.4).

Carbon

The equation used to derive $\delta^{13}C$ is provided below. For example, a sample with $\delta^{13}C$ of 35‰ has 3.5% more ^{13}C than the reference material (which is a fossil carbonate mineral known as Vienna Pee Dee Belemite), and therefore is described as relatively enriched in ^{13}C.

$$\delta^{13}\mathrm{C} = \left(\frac{(C^{13}/C^{12})_{sample} - (C^{13}/C^{12})_{reference}}{(C^{13}/C^{12})_{reference}}\right) \times 1000‰ \qquad \text{(Equation 2)}$$

Measured $\delta^{13}C$ ranges were compared between species. Where isotopic signatures of prey and consumer overlapped, the taxa were identified as potential prey (Hecky and Hesslein 1995; Post 2002). A *potential* prey item does not necessarily indicate a biologically *likely* item. Biologically unlikely results were identified in the integrated analysis (Section 3.2.4). Isotopic signatures were also used to distinguish between littoral and pelagic feeding within Dinosaur Reservoir, with the more depleted $\delta^{13}C$ suggesting pelagic feeding (Post 2002; France 1995).

Trophic Position

Estimating trophic position for a species from $\delta^{15}N$ data requires an estimate of the “baseline” nitrogen isotopic signature for the food web. Primary producers and detritus that represent the base of aquatic food webs have high temporal and spatial variation in $\delta^{15}N$ that can be challenging to characterize (Post 2002; Cabana and Rasmussen 1996). Longer lived species produce more consistent and reliable measures of $\delta^{15}N$ compared to shorter lived species and detritus. Therefore, Post (2002) recommended using a long-lived primary consumer to estimate the baseline $\delta^{15}N$ of a food web.

Trophic position was calculated for each species based on a one-source food web, where there is only one estimate of $\delta^{15}N$ for the base of the food web, using Equation 3:

$$\text{Trophic Position} = \lambda + \frac{\delta^{15}N_{SC} - \delta^{15}N_{base}}{\Delta_n} \quad \text{(Equation 3)}$$

where λ indicates the trophic position of species used to estimate $\delta^{15}N_{base}$; $\delta^{15}N_{SC}$ is the mean nitrogen isotope ratio of the secondary consumer; $\delta^{15}N_{base}$ is the mean nitrogen isotope ratio of the base of the food web; and, Δ_n is the $\delta^{15}N$ enrichment per trophic level.

Trophic position was also calculated based on a two-source food web, using Equation 4:

$$\text{Trophic Position} = \lambda + \frac{\delta^{15}N_{SC} - [\delta^{15}N_{base1} \times \propto + \delta^{15}N_{base2} \times (1 - \propto)]}{\Delta_n} \quad \text{(Equation 4)}$$

where $\delta^{15}N_{base1}$ is the mean nitrogen isotope ratio of one base of the food web and $\delta^{15}N_{base2}$ is the mean nitrogen isotope ratio of the second base of the food web.

For Equation 4, the variable, α, represents the proportion of consumed nitrogen derived from the first base of the food web, and can be estimated according to Equation 5:

$$\propto = \frac{\delta^{13}C_{SC} - \delta^{13}C_{base2}}{\delta^{13}C_{base1} - \delta^{13}C_{base2}} \quad \text{(Equation 5)}$$

The following assumptions were applied to these equations:

1) The base of the food web was estimated using available data from primary consumers: gastropods, mayflies and caddisflies.

2) The trophic position of the primary consumer, λ, was assumed to be 2; however, there is some uncertainty with this estimation because mayflies are known to be omnivores, and may be more accurately represented by a trophic position greater than 2 (Vander Zanden et al. 1997).

3) The enrichment per trophic level, Δ_n, was assumed to be 3.4‰. This assumption is based on Post (2002) and Perkins et al. (2004), but the authors acknowledge that nitrogen isotope enrichment can range from 2.5‰ to 4‰ and, in some studies, deviate from this range. Post (2002) concluded that uncertainty in Δ_n contributed less to the overall uncertainty and variability in calculating trophic position compared to the estimation of $\delta^{15}N_{base}$.

To account for the uncertainty in the base of the food web, trophic position was calculated under four different scenarios where each scenario represents a different estimation of the $\delta^{15}N$ at the base of the food web. The four separate scenarios are described in Table 1 and vary based on assumptions about the base of food chain. These scenarios vary based on three main factors:

- Waterbody: Scenario A only includes data collected in Dinosaur Reservoir, whereas Scenarios B, C, and D only include data collected in the Peace River.

- Species used to estimate $\delta^{15}N_{base}$: Scenarios A and B only include gastropods, whereas Scenarios C and D include gastropods, mayflies, and caddisflies[1].
- Food web structure: Scenarios A, B, and C calculate trophic position based on a one-source food, whereas Scenario D calculates trophic position based on a two-source food web. In scenario D, $\delta^{15}N_{base}$ of the first source (i.e., base of food web) was estimated from gastropods and $\delta^{15}N_{base}$ of the second source was estimated from mayflies and caddisflies.

Table 2: Summary of variables used in trophic position calculations for the Peace River, 2020.

Scenario	Waterbody	Species use to Estimate $\delta^{15}N_{base}$	Food Web Structure[a]	Formula[b]
A	Dinosaur Reservoir	Dinosaur Reservoir gastropods only	One-source	$\lambda + (\delta^{15}N_{SC} - \delta^{15}N_{base}) / \Delta^{15}N$
B	Peace River	Peace River gastropods only	One-source	$\lambda + (\delta^{15}N_{SC} - \delta^{15}N_{base}) / \Delta^{15}N$
C	Peace River	Peace River gastropods, mayflies, caddisflies	One-source	$\lambda + (\delta^{15}N_{SC} - \delta^{15}N_{base}) / \Delta^{15}N$
D	Peace River	Peace River gastropods, mayflies, caddisflies	Two-source (Gastropods; Mayflies & Caddisflies)	$\lambda + (\delta^{15}N_{SC} – [\delta^{15}N_{baseGA} * \alpha + \delta^{15}N_{baseET} * (1 – \alpha)]) / \Delta^{15}N$

[a] One-source uses one estimate of $\delta^{15}N_{base}$ and two-source uses two separate estimates of $\delta^{15}N_{base}$.
[b] λ = trophic position of species used to estimate $\delta^{15}N_{base}$ (assumed to be 2); $\delta^{15}N_{SC}$ = mean nitrogen isotope ratio of the secondary consumer; $\delta^{15}N_{base}$ = mean nitrogen isotope ratio of the base of the food web; $\Delta^{15}N$ = $\delta^{15}N$ enrichment per trophic level (assumed to be 3.4‰); $\delta^{15}N_{baseGA}$ = mean nitrogen isotope ratio of gastropods; $\delta^{15}N_{baseET}$ = mean nitrogen isotope ratio of mayflies and caddisflies; α = proportion of consumed nitrogen derived from gastropods, calculated as $(\delta^{13}C_{secondary\ consumer} – \delta^{13}C_{baseET}) / (\delta^{13}C_{baseGA} – \delta^{13}C_{baseET})$ where $\delta^{13}C_{baseGA}$ = mean carbon isotope ratio of gastropods and $\delta^{13}C_{baseET}$ = mean carbon isotope ratio of mayflies and caddisflies.

3.2.2 Mercury

Tissue mercury data were available for a subsample of fish in the “BC Hydro Peace River Mercury and SIA Database version 2020.1.20” (Golder 2020), which allowed for a preliminary assessment of bioaccumulation. Tissue samples were collected as either biopsy plug or fillet. Where data were provided as wet weight, data were converted to dry weight using the measured percent moisture for the respective tissue sample. Tissue mercury concentrations were plotted against measured $\delta^{15}N$ and trends were compared among species.

3.2.3 Stomach Contents

Stomach content samples from 2010, 2011, 2017, and 2018 collected from six different fish species in Dinosaur Reservoir and Peace River were included in the stomach content analyses (Table 3). These data were provided as total abundance of each taxonomic group and biomass data were not provided. Detailed methods regarding the collection of stomach contents are found in Ecoscape (2018).

[1] The only invertebrates collected from Dinosaur Reservoir were gastropods; therefore, additional scenarios to estimate $\delta^{15}N$ at the base of the food web are not possible for Dinosaur Reservoir.

The relative abundance of major taxonomic groups was calculated as a percent of the total abundance (i.e., the absolute number of individuals found in each fish stomach sample). Invertebrates groups or species with no aquatic life stage were grouped under a single category called "terrestrial".

Table 3: Number of stomach content samples collected from Dinosaur Reservoir and the Peace River and included in stomach content analyses.

Location	Year	Species	Number of Samples
Dinosaur Reservoir	2010	Kokanee	119
		Mountain Whitefish	56
		Rainbow Trout	141
	2011	Longnose Sucker	79
		Lake Trout	26
		Mountain Whitefish	206
		Rainbow Trout	175
	2017	Arctic Grayling	119
		Longnose Sucker	12
		Mountain Whitefish	186
		Rainbow Trout	49
	2018	Arctic Grayling	49
		Mountain Whitefish	163
		Rainbow Trout	35
Peace River	2010	Arctic Grayling	100
		Kokanee	26
		Mountain Whitefish	146
		Rainbow Trout	46
	2011	Kokanee	113
		Longnose Sucker	27
		Mountain Whitefish	179
		Rainbow Trout	208
	2017	Arctic Grayling	185
		Mountain Whitefish	112
		Rainbow Trout	190
	2018	Arctic Grayling	191
		Mountain Whitefish	240
		Rainbow Trout	177

3.2.4 Integrated Food Web Analysis

Results from the stable isotope analysis were used to identify *potential* prey items which were compared with stomach content analysis results and published information on dietary habits (McPhail 2007) to characterize biologically *likely* prey items for each species. First, the results from $\delta^{15}N$ analysis were used to identify all potential prey items. Species identified by the $\delta^{13}C$ analysis as unlikely to be in the same food web were then

excluded as potential prey items. Stomach contents were used to confirm or add biologically likely prey species. Literature (predominantly McPhail 2007 unless otherwise cited) was reviewed to categorize fish as piscivorous, non-piscivorous, or occasionally piscivorous, furthering filtering biologically unlikely prey items.

4.0 RESULTS

4.1 Stable Isotope Analysis

A summary of stable isotope data collected in Dinosaur Reservoir and the Peace River is provided in Table 4. Mean $\delta^{15}N$ ranged from 8.5‰ to 12‰ in fish species in Dinosaur Reservoir and 8.0‰ to 11‰ in the Peace River. Gastropods were the only invertebrates collected in Dinosaur Reservoir, for which mean $\delta^{15}N$ was 3.3‰. Gastropods, mayflies (order Ephemeroptera) and caddisflies (order Trichoptera) were collected in the Peace River, and mean $\delta^{15}N$ of these invertebrates was 4.1‰, 2.4‰, and 2.2‰, respectively. Mean $\delta^{13}C$ ranged from -27‰ to -24‰ in fish species in Dinosaur Reservoir and -25‰ to -29‰ in the Peace River. For gastropods, mean $\delta^{13}C$ in Dinosaur Reservoir was -25‰, whereas mean $\delta^{13}C$ for gastropods, mayflies, and caddisflies in the Peace River were -26‰, -28‰, and -31‰, respectively.

Fewer species were collected from Dinosaur Reservoir (5 fish species; 1 invertebrate species) compared to the Peace River (11 fish species; 3 invertebrate species). Samples sizes were small (fewer than 10 samples) for gastropods in Dinosaur Reservoir (n = 4) and for the following species in the Peace River: Arctic Grayling (n = 3), Lake Trout (n = 3), gastropods (n = 7), and caddisflies (n = 6). The discrepancy in sample sizes may in part reflect the greater sampling effort conducted across nine separate sections of the Peace River (Attachment 1, Figure 1A).

Biplots depicting $\delta^{15}N$ and $\delta^{13}C$ are provided in Figure 1 for Dinosaur Reservoir and Figure 2 for the Peace River. In both waterbodies, invertebrate species group separately from fish species. In Dinosaur Reservoir, fish species appear to be further separated into two distinct groupings, with Lake Trout and Bull Trout associated with higher $\delta^{15}N$ (higher trophic level) and lower $\delta^{13}C$ (feeding on a more ^{13}C-depleted food web) compared to Longnose Sucker (*Catostomus catostomus*), Mountain Whitefish, and Rainbow Trout. In the Peace River, there is no apparent separation of fish species on the $\delta^{13}C$ axis, suggesting that all fish share the same basal resource, and a continuous range of $\delta^{15}N$, indicating a range of piscivory, partial piscivory, and non-piscivory.

Table 4: Summary of stable isotope data collected from various fish species in Dinosaur Reservoir and the Peace River.

Species	Number of Samples	$\delta^{15}N$ (‰)[a]		$\delta^{13}C$ (‰)[a]	
		Mean	Standard Error	Mean	Standard Error
Dinosaur Reservoir					
Bull Trout	15	11	0.14	-34	0.51
Lake Trout	30	12	0.086	-32	0.39
Longnose Sucker	12	9.3	0.18	-28	0.30
Mountain Whitefish	26	8.7	0.13	-27	0.24
Rainbow Trout	10	8.5	0.11	-26	0.26
Gastropods	4	3.3	0.44	-25	0.71
Peace River					
Arctic Grayling	4	7.8	0.21	-28	0.15
Bull Trout	107	10.4	0.07	-29	0.23
Burbot	22	10.2	0.19	-27	0.27
Goldeye	25	9.1	0.11	-26	0.11
Lake Trout	4	11.9	0.72	-28	0.050
Longnose Sucker	199	7.3	0.08	-28	0.13
Mountain Whitefish	251	8.6	0.06	-29	0.12
Northern Pike	41	9.5	0.15	-27	0.13
Rainbow Trout	40	8.9	0.12	-28	0.43
Redside Shiner	12	8.2	0.08	-26	0.14
Walleye	92	10.7	0.07	-26	0.080
Mayflies	22	2.4	0.25	-28	0.42
Gastropods	7	4.1	0.32	-26	0.42
Caddisflies	6	2.2	0.39	-31	0.44

[a] $\delta^{15}N$ = nitrogen isotope ratio; $\delta^{13}C$ = carbon isotope ratio.

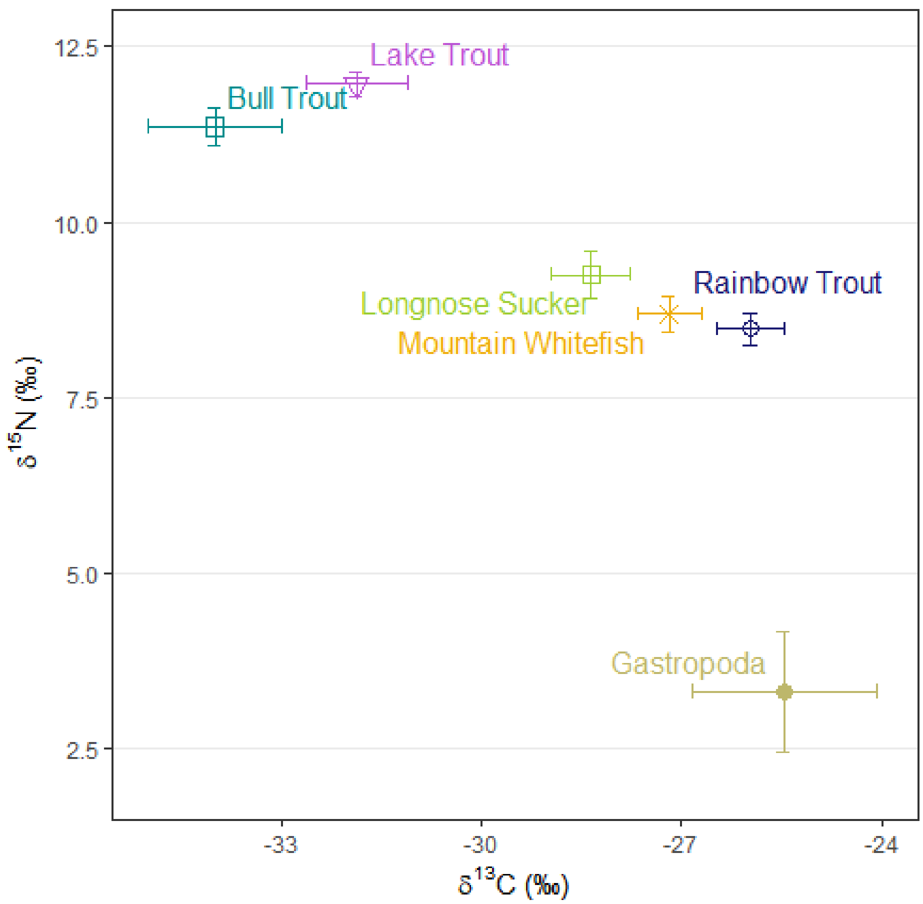

Figure 1: Stable isotope biplot for various fish species in Dinosaur Reservoir. Symbols represent mean values and error bars represent standard error.

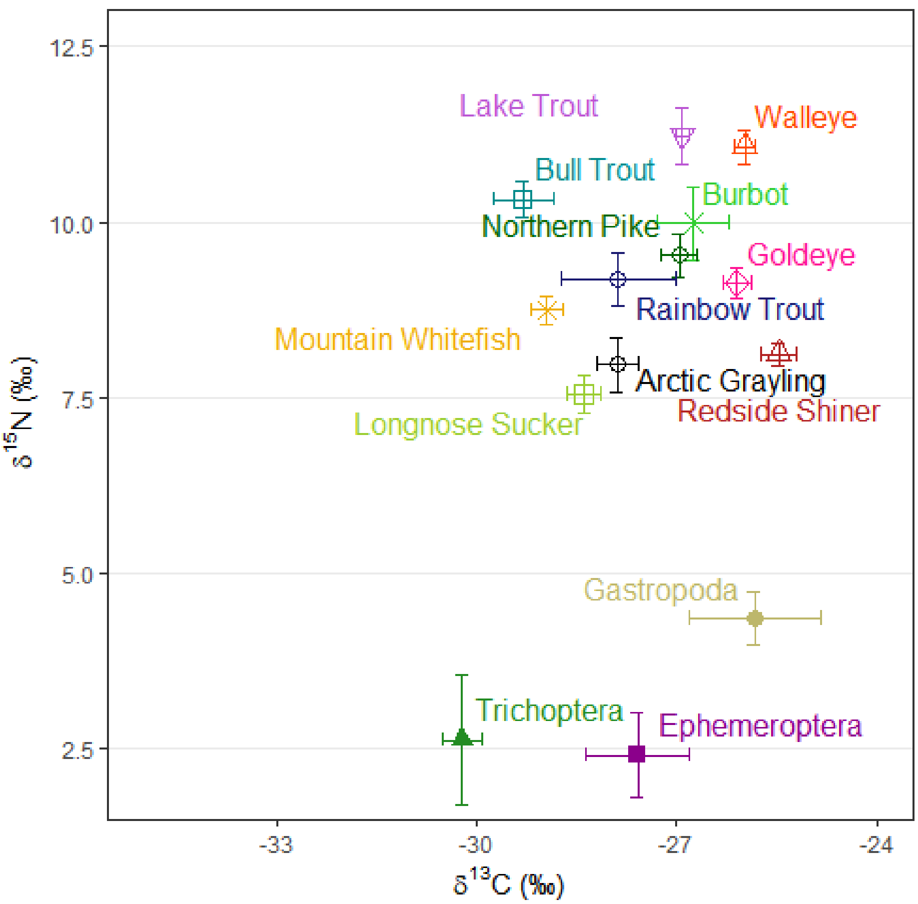

Figure 2: Stable isotope biplot for various fish species in the Peace River. Symbols represent mean values and error bars represent standard error.

4.1.1 Nitrogen

Stable isotope ratios of nitrogen ($\delta^{15}N$) for fish and invertebrate species collected in Dinosaur Reservoir and the Peace River are presented in Figure 3. Variability in standard error was observed across locations and may reflect heterogeneity of diet or the low sample size for given species and locations.

As expected, mean $\delta^{15}N$ was greater for fish species (consumers) compared to invertebrate species (prey), and greater for piscivorous fish (e.g., Lake Trout, Bull Trout, Burbot, Northern Pike, and Walleye) compared to non-piscivorous fish (e.g., Longnose Sucker, Mountain Whitefish).

For all fish species sampled in both waterbodies, $\delta^{15}N$ in Dinosaur Reservoir was consistent with the range observed across sections in the Peace River. However, estimating trophic positioning requires a reliable estimate of the $\delta^{15}N$ of the base of the food web, represented in this dataset by gastropods, mayflies, and caddisflies. There was greater variability in mayflies compared to gastropods, consistent with results presented by Post (2002), and has implications for uncertainty in determining the trophic position of each species, which is discussed further below (Section 4.1.3).

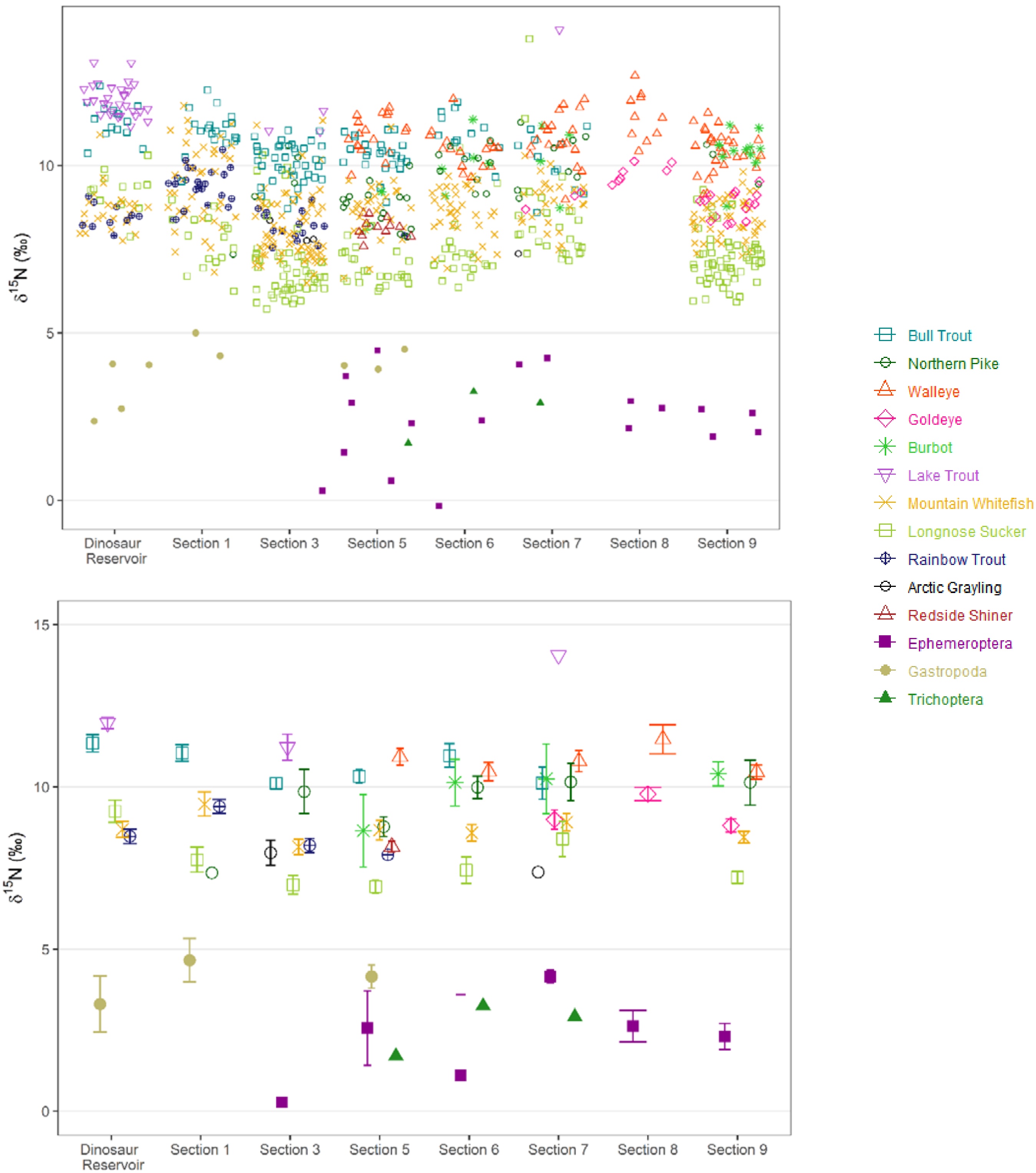

Figure 3: Isotopic nitrogen ratios in Dinosaur Reservoir and in the Peace River by section. Data are presented as individual data points in the top panel and as mean values with standard error in the bottom panel.

Potential prey items were identified by an overlap in the $\delta^{15}N$ range between prey item and consumer and are summarized in Table 5. It is important to note that *potential* species of prey are not necessarily *likely* species of prey. Likelihood of prey items is further expanded in Section 5.0 where these results were filtered to exclude biologically unrealistic pairings.

Fewer potential prey items were identified using $\delta^{15}N$ range for consumers in Dinosaur Reservoir compared to the Peace River, likely due to the low number of fish species sampled in Dinosaur Reservoir. The only invertebrates collected in Dinosaur Reservoir were gastropods, which were the only prey item identified for Rainbow Trout, an occasionally piscivorous species (McPhail 2007). Both fish and gastropods were identified as potential prey items for Longnose Sucker and Mountain Whitefish, although fish are unlikely prey for these non-piscivorous species. Only fish were identified as potential prey for Lake Trout and Bull Trout, two piscivorous species.

In the Peace River, fish were the only identified prey of Lake Trout, and Walleye, species known to be piscivorous (McPhail 2007). Both fish and invertebrates were identified as potential prey for all other fish species, although fish are unlikely prey for Redside Shiner (*Richardsonius balteatus*), Mountain Whitefish, Arctic Grayling, and Longnose Sucker, which are non-piscivorous species (McPhail 2007). Longnose Sucker was identified as a potential prey item for all fish species, which may be driven by the large within-species range in $\delta^{15}N$ for Longnose Sucker.

Table 5: Potential prey items identified by isotopic nitrogen signatures analyzed from samples collected in Dinosaur Reservoir and the Peace River.

Consumer	Potential Prey Items													
	GA	EP	TR	LT	WP	BT	BB	NP	GE	RB	MW	RSC	AG	LSU
Dinosaur Reservoir[a]														
LT						x[b]				x	x			x
BT										x	x			x
RB	x													
MW	x									x	x			x
LSU[c]	x										x			
Peace River														
LT						x	x	x	x	x	x	x	x	x
WP					x	x	x	x	x	x	x	x	x	x
BT	x				x	x	x	x	x	x	x	x	x	x
BB	x	x				x	x		x	x	x	x	x	x
NP	x						x		x		x	x	x	x
GE	x	x									x	x		x
RB	x	x									x	x		x
MW	x	x	x			x	x	x	x	x	x	x	x	x
RSC	x	x												x
AG	x	x												x
LSU[1]	x	x	x								x			x

Note: *Potential* species of prey are not necessarily *likely* species of prey. Likelihood of prey items is further expanded in Section 5.0.
[a] Shading indicates that the species was not collected in Dinosaur Reservoir.
[b] 'X' indicates a potential prey item, identified by overlapping $\delta^{15}N$ ranges by 2.5‰ – 4‰.
[c] Two outliers (13.7‰ and 11.4‰) were visually identified and removed for calculation of potential prey items for Longnose Sucker.
GA = gastropods, EP = mayflies; TR = caddisflies, LT = Lake Trout, WP = Walleye; BT = Bull Trout; NP = Northern Pike; GE = Goldeye; RB = Rainbow Trout; MW = Mountain Whitefish; RSC = Redside Shiner; AG = Arctic Grayling; LSU = Longnose Sucker.

4.1.2 Carbon

Stable isotope ratios of carbon ($\delta^{13}C$) for fish and invertebrate species collected in Dinosaur Reservoir and the Peace River are presented in Figure 4. Variability in standard error was observed across locations and may reflect heterogeneity of diet or low sample sizes for given species and locations.

Within-species $\delta^{13}C$ was consistent between Dinosaur Reservoir and the Peace River, with the exception of Bull Trout and Lake Trout, both of which had depleted mean $\delta^{13}C$ in Dinosaur Reservoir compared to the Peace River. In Dinosaur Reservoir, Lake Trout and Bull Trout had depleted (i.e., less enriched) $\delta^{13}C$ compared to other fish species. Within lakes, pelagic food webs are typically $\delta^{13}C$ depleted when compared to benthic food webs (Post 2002; France 1995). This suggests that Lake Trout and Bull Trout in Dinosaur Reservoir may be feeding on pelagic prey while Longnose Sucker, Mountain Whitefish, and Rainbow Trout may be feeding on benthic food sources.

There are no invertebrates with similar $\delta^{13}C$ ranges as Lake Trout and Bull Trout in the Dinosaur Reservoir, which is consistent with the observation that prey items for Lake Trout and Bull Trout were not collected in this study. Further, this suggests that the gastropods collected in this study do not reflect the base of the food web for Lake Trout and Bull Trout. This has implications for uncertainty in the trophic position calculations, as discussed in Section 4.1.3.

Potential prey items were identified by an overlap in the $\delta^{13}C$ range between prey item and consumer and are summarized in Table 6. It is important to note that *potential* species of prey are not necessarily *likely* species of prey. Likelihood of prey items is further expanded on in Section 5.0 where these results were filtered to exclude biologically unrealistic pairings.

A greater number of potential prey items was identified by carbon signatures compared to nitrogen signatures; however, overlapping $\delta^{13}C$ ranges do not necessarily indicate potential prey items, whereas the absence of overlapping $\delta^{13}C$ ranges is a reliable indicator that a species is an unlikely prey item. Overlapping $\delta^{13}C$ ranges may indicate species are feeding within the same food web and not necessarily indicate a consumer-prey relationship. Fewer potential prey items were identified for consumers in Dinosaur Reservoir compared to the Peace River, likely due to the lower number in species sampled in Dinosaur Reservoir.

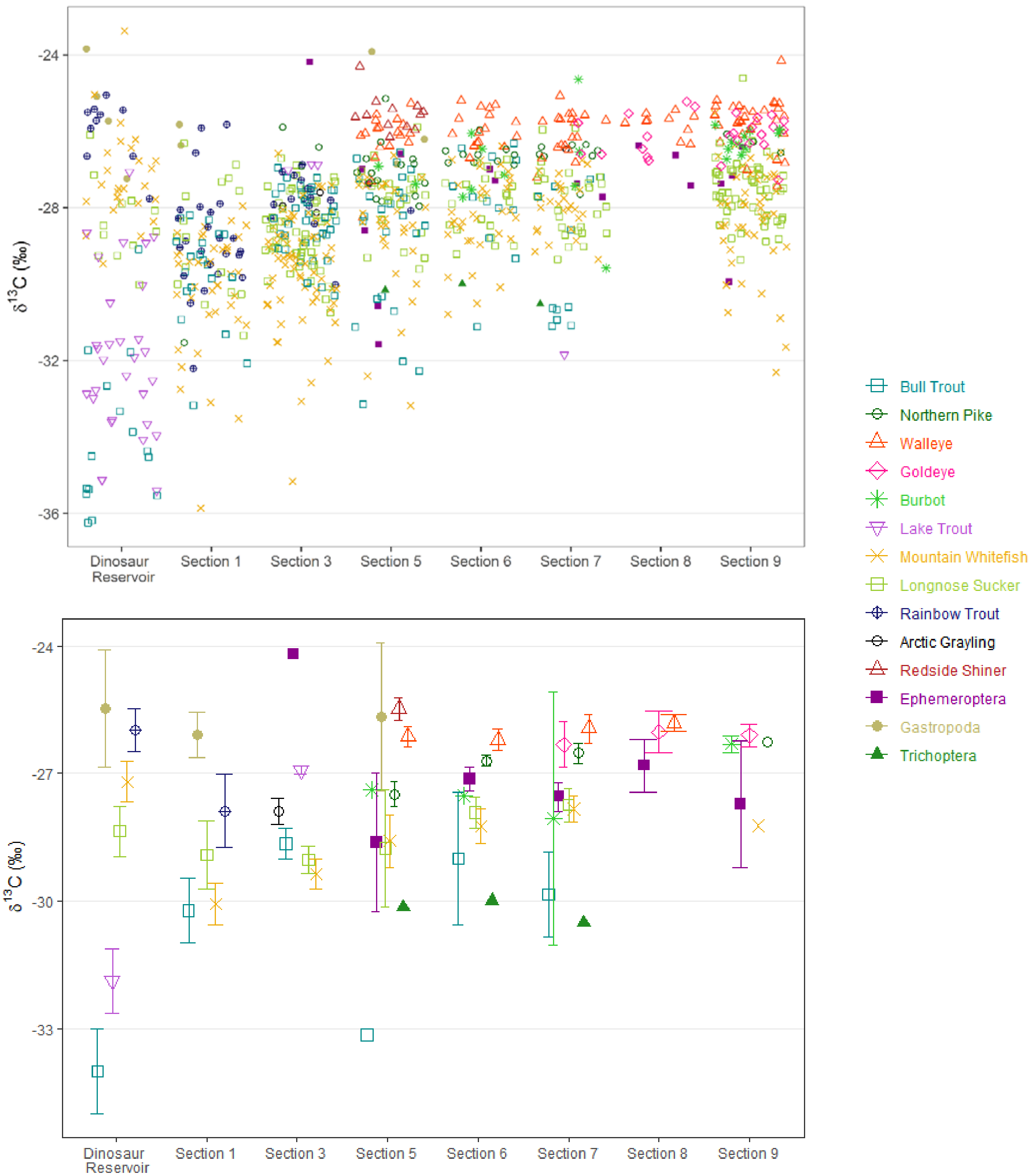

Figure 4: Isotopic carbon signature for various fish and invertebrate species in Dinosaur Reservoir and the Peace River. Data are presented as individual data points in the top panel and as mean values with standard error in the bottom panel.

Table 6: Potential prey items identified by overlapping δ^{13}C ranges in Dinosaur Reservoir and the Peace River.

Consumer	Potential Prey Items													
	GA	EP	TR	LT	WP	BT	BB	NP	GE	RB	MW	RSC	AG	LSU
Dinosaur Reservoir[a]														
LT	X[b]			x		x				x	x			x
BT				x		x					x			x
RB	x			x						x	x			x
MW	x			x		x				x	x			x
LSU	x			x		x				x	x			x
Peace River														
LT	x	x		x			x	x	x	x	x			x
WP	x	x			x		x	x	x	x	x	x		x
BT		x	x			x	x	x	x	x	x		x	x
BB	x	x	x	x	x	x	x	x	x	x	x	x	x	x
NP	x	x		x	x	x	x	x	x	x	x		x	x
GE	x	x		x	x	x	x	x	x	x	x	x		x
RB	x	x	x	x	x	x	x	x	x	x	x	x	x	x
MW	x	x	x	x	x	x	x	x	x	x	x		x	x
RSC	x	x			x		x		x	x		x		x
AG		x				x	x	x		x	x		x	x
LSU	x	x	x	x	x	x	x	x	x	x	x	x	x	x

Note: It is important to note that *potential* species of prey are not necessarily *likely* species of prey. Likelihood of prey items is further expanded in Section 5.0.
[a] Shading indicates that this species was not collected in Dinosaur Reservoir.
[b] 'X' indicates a potential prey item, identified by overlapping δ^{13}C ranges. Overlapping δ13C ranges may indicate species are feeding within the same food web and not necessarily indicate a consumer-prey relationship
GA = gastropods, EP = mayflies; TR = caddisflies, LT = Lake Trout, WP = Walleye; BT = Bull Trout; NP = Northern Pike; GE = Goldeye; RB = Rainbow Trout; MW = Mountain Whitefish; RSC = Redside Shiner; AG = Arctic Grayling; LSU = Longnose Sucker.

4.1.3 Trophic Position

As identified in Section 4.1.1, δ^{15}N measured in mayfly samples was variable, and these samples comprise the majority of samples expected to represent the base of the food web. Trophic positions were calculated under four food web base scenarios to account for this uncertainty and are presented in Table 7.

Table 7: Trophic position calculated under four scenarios for various fish species in Dinosaur Reservoir and the Peace River.

Species	Trophic Position (± Standard Error)[a,b]			
	Scenario A	Scenario B	Scenario C	Scenario D
Waterbody	Dinosaur Reservoir	Peace River	Peace River	Peace River
Base of Food Web[c]	One-source (GA)	One-source (GA)	One-source (GA, EP, TR)	Two-source (GA & EP, TR)
$\delta^{15}N_{base}$ (‰)	3.3	4.1	2.7	2.7
Lake Trout	4.5 ± 0.1	4.1 ± 0.1	4.5 ± 0.1	4.3 ± 0.2
Walleye	-	4.1 ± 0.1	4.5 ± 0.1	4.2 ± 0.1
Bull Trout	4.4 ± 0.1	3.8 ± 0.1	4.2 ± 0.1	4.0 ± 0.1
Burbot	-	3.7 ± 0.1	4.1 ± 0.1	3.9 ± 0.3
Northern Pike	-	3.6 ± 0.1	4.0 ± 0.1	3.8 ± 0.2
Goldeye	-	3.5 ± 0.1	3.9 ± 0.1	3.7 ± 0.1
Rainbow Trout	3.5 ± 0.1	3.5 ± 0.1	3.9 ± 0.1	3.7 ± 0.2
Mountain Whitefish	3.6 ± 0.1	3.4 ± 0.1	3.8 ± 0.1	3.5 ± 0.1
Redside Shiner	-	3.2 ± 0.1	3.6 ± 0.1	3.4 ± 0.1
Arctic Grayling	-	3.1 ± 0.1	3.6 ± 0.1	3.3 ± 0.2
Longnose Sucker	3.7 ± 0.1	3.0 ± 0.1	3.4 ± 0.1	4.2 ± 0.2

[a] Trophic position of a one-source food web = $\lambda + (\delta^{15}N_{secondary\ consumer} - \delta^{15}N_{base}) / \Delta N$; where:

- λ = trophic position of species used to estimate $\delta^{15}N_{base}$ (assumed to be 2)
- $\delta^{15}N_{secondary\ consumer}$ = mean nitrogen isotope ratio of the species
- $\delta^{15}N_{base}$ = mean nitrogen isotope ratio of the base of the food web
- ΔN = is the $\delta^{15}N$ enrichment per trophic level (assumed to be 3.4‰)

[b] Trophic position of a two-source food web = $\lambda + (\delta^{15}N_{secondary\ consumer} - [\delta^{15}N_{baseGA} * \alpha + \delta^{15}N_{baseET} * (1 - \alpha)]) / \Delta N$;

- $\delta^{15}N_{baseGA}$ = mean nitrogen isotope ratio of gastropods
- $\delta^{15}N_{baseET}$ = mean nitrogen isotope ratio of mayflies and caddisflies
- α = proportion of consumed nitrogen derived from gastropods, calculated as $(\delta^{13}C_{secondary\ consumer} - \delta^{13}C_{baseET}) / (\delta^{13}C_{baseGA} - \delta^{13}C_{baseET})$; where:
  - $\delta^{13}C_{baseGA}$ = mean carbon isotope ratio of gastropods
  - $\delta^{13}C_{baseET}$ = mean carbon isotope ratio of mayflies and

[c] One-source uses one estimate of $\delta^{15}N_{base}$ and two-source uses two separate estimates of $\delta^{15}N_{base}$.

$\delta^{15}N$ = nitrogen isotope ratio; ‰ = permil; GA = gastropods; EP = mayflies; TR = caddisflies

The relative trophic positions of species in Dinosaur Reservoir and the Peace River across the four food web base scenarios were the same, with Lake Trout and Walleye occupying the highest trophic positions and Longnose Sucker and Arctic Grayling occupying the lowest trophic positions. Overall, trophic positions were higher when calculated under Scenarios C and D, which were the scenarios that calculated the base of the food web using mayflies and caddisflies. Fish species collected from both Dinosaur Reservoir and the Peace River (i.e., Lake Trout, Bull Trout, Rainbow Trout, Mountain Whitefish, and Longnose Sucker) had higher trophic positions in Dinosaur Reservoir when compared to the Peace River, driven by the lower estimate of $\delta^{15}N_{base}$. As identified by the carbon analyses (see Section 4.1.2), this dataset does not capture the base of the food web for Lake Trout and Bull Trout. Therefore, the corresponding trophic position estimates have greater uncertainty for these species compared to other fish species.

As expected, fish known to be piscivorous (i.e., Lake Trout, Walleye, Bull Trout, Burbot, and Northern Pike; McPhail 2007) had the highest trophic positions. Fish known to be non-piscivorous (i.e., Redside Shiner, Longnose Sucker; McPhail 2007) had the lowest trophic positions. Other authors indicate Arctic Grayling, Goldeye, Rainbow Trout, and Mountain Whitefish can be piscivorous depending on food availability (McPhail 2007). In the Peace River, Arctic Grayling are expected to be non-piscivorous, as they occupy trophic positions between Redside Shiner and Longnose Sucker. Goldeye, Rainbow Trout, and Mountain Whitefish occupy trophic positions between the piscivorous and non-piscivorous fish and therefore the piscivorous nature of these fish in Dinosaur Reservoir and the Peace River remains uncertain.

4.2 Mercury Analysis

A summary of tissue mercury data collected for a subset of the fish collected in Dinosaur Reservoir and the Peace River is provided in Table 8. Of the individual fish for which mercury data were available, a subset had paired stable isotope data. There were fewer than five individual samples with paired data for Longnose Sucker in Dinosaur Reservoir and Lake Trout and Arctic Grayling in the Peace River.

Table 8: Summary of tissue mercury data collected from various fish species in Dinosaur Reservoir and the Peace River.

Species	Number of Samples		Mercury Concentration (mg/kg dw)	
	Total Number of Fish	Number of Fish with Paired SIA data	Mean	Standard Error
Dinosaur Reservoir				
Bull Trout	16	15	0.51	0.09
Lake Trout	20	20	0.42	0.03
Longnose Sucker	1	1	0.92	-
Mountain Whitefish	15	15	0.20	0.02
Peace River				
Arctic Grayling	4	3	0.14	0.04
Bull Trout	159	105	0.45	0.03
Burbot	25	21	0.59	0.06
Goldeye	17	14	1.19	0.06
Lake Trout	4	4	0.66	0.13
Longnose Sucker	242	148	0.34	0.02
Mountain Whitefish	277	195	0.22	0.01
Northern Pike	48	36	0.68	0.11
Rainbow Trout	50	25	0.17	0.03
Redside Shiner	12	10	0.22	0.01
Walleye	130	86	1.11	0.06

The relationship between tissue mercury concentrations and paired δ^{15}N measurements is presented in Figure 5 for Dinosaur Reservoir and Figure 6 for the Peace River. Spatial trends in mercury across Dinosaur Reservoir and Peace River are presented in Figure 7. No paired data were available for Section 8 in the Peace River.

Mercury concentration generally increased with increasing δ^{15}N enrichment, likely reflecting the bioaccumulation of mercury across trophic levels (Figure 6, Figure 7). Mercury increased to a lesser extent with increased δ^{15}N in Lake Trout from Dinosaur Reservoir and Longnose Sucker from the Peace River, compared to other fish species. Compared to fish species with similar δ^{15}N, Goldeye had higher levels of Mercury. This may reflect increased bioaccumulation or uptake of mercury in Goldeye or indicate that Goldeye feed on different prey items compared to other fish sampled.

There were higher tissue mercury concentrations in Sections 3 to 9 relative to Dinosaur Reservoir and Section 1. However, this is likely due to an increased number of Northern Pike, Goldeye, and Walleye sampled in these sections, as fish of these species had higher δ^{15}N and mercury concentrations.

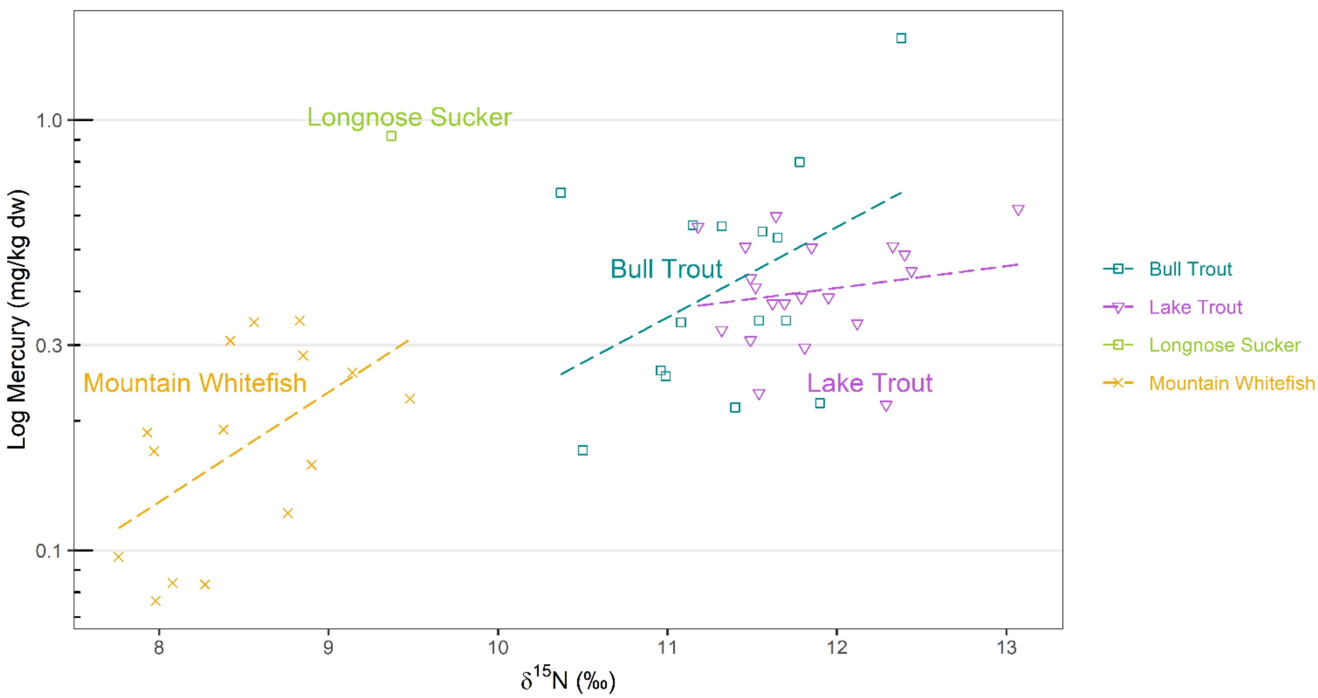

Figure 5: Mercury and nitrogen relationships for various fish species in Dinosaur Reservoir. Dashed lines represent the line of best fit. Mercury in presented as dry weight.

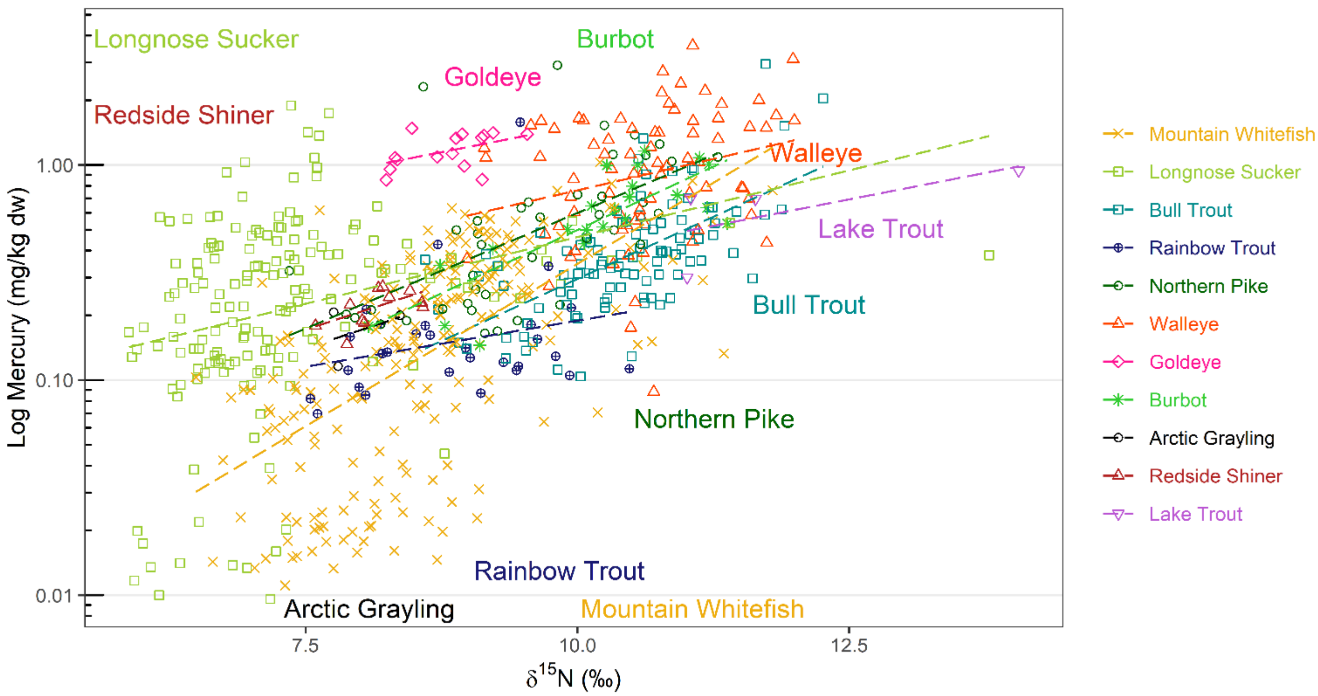

Figure 6: Mercury and nitrogen relationship for various fish species in the Peace River. Dashed lines represent the line of best fit. Mercury is presented in dry weight.

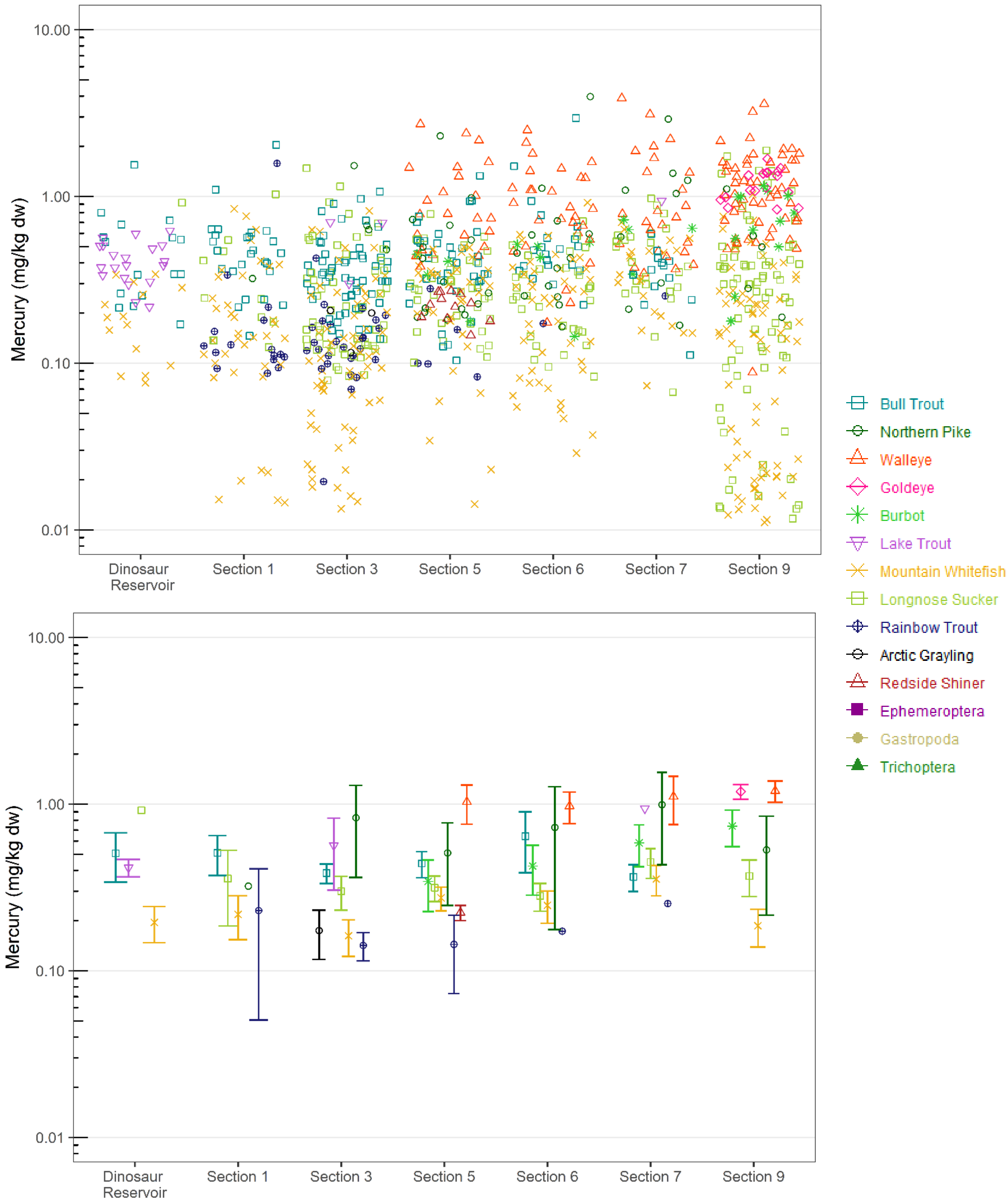

Figure 7: Mercury concentrations in various fish species in Dinosaur Reservoir and the Peace River. Mercury is presented in dry weight.

4.3 Stomach Contents

A summary of the relative abundance of major taxa found in fish collected in Dinosaur Reservoir and the Peace River is provided in Table 9, Table 10, Figure 8 and Figure 9. Stomach contents were available for Arctic Grayling, Kokanee, Mountain Whitefish, Longnose Sucker, Lake Trout, Mountain Whitefish, and Rainbow Trout. Stomach content analysis was limited to invertebrates and therefore does not inform on fish species of prey. Volumetric or weight data were not available for the prey items, which presents uncertainty in assessing prey importance, as fewer large items may provide greater nutritional content compared to several small items.

Across fish species, the following invertebrates were found in fish stomach contents in both Dinosaur Reservoir and the Peace River: copepods (Order: Copepoda), caddisflies, terrestrial insects, mayflies, and true flies (Order: Diptera). Leeches (Order: Hirudinea) and true bugs (Order: Hemiptera) were found only in the Peace River and seed shrimp (Order: Ostracoda) were found only in Dinosaur Reservoir.

Lake Trout were the only piscivorous fish for which stomach content data were available, with 26 samples collected from Dinosaur Reservoir in 2011 comprised of primarily true flies (Ecoscape 2018). Stomach content data available were limited to invertebrates and this result does not reflect the piscivorous component of the Lake Trout diet. Mountain Whitefish sampled from the Peace River (2010, 2011, 2017, and 2018) consumed predominantly true flies and caddisflies and consumed a greater proportion of true flies in Dinosaur Reservoir. Arctic Grayling in the Dinosaur Reservoir (2017 and 2018) consumed predominantly terrestrial insects, mayflies, and caddisflies, and Arctic Grayling in the Peace River (2017 and 2018) consumed predominantly true flies and mayflies. Rainbow Trout in the Dinosaur Reservoir (2010, 2011, 2017, 2018) consumed predominantly mayflies or true flies, and Rainbow Trout in the Peace River (2010, 2011, 2017, and 2018) consumed predominantly mayflies. Longnose Sucker in the Dinosaur Reservoir consumed predominantly true flies in 2011 and mayflies in 2017, and Longnose Sucker in the Peace River consumed predominantly true flies.

Table 9: Relative abundance of prey items identified in the stomach contents of fish captured in Dinosaur Reservoir.

Species	Year	Relative Abundance (%)							
		True Flies	Water Fleas	Mayflies	Seed Shrimp	Terrestrial	Caddis-flies	Copepods	Other Taxa
Arctic Grayling	2017	2.4	0	21	0	67	2.0	0	7.3
	2018	5.3	0	31	0	8.4	32	0	24
Kokanee	2010	2.0	28	0.0087	0	0.32	0.81	69	0.0087
Longnose Sucker	2011	67.2	0	0	31	0.04	0.094	0.21	1.0
	2017	2.2	0	0.86	0	0	94	0	3.1
Lake Trout	2011	99	0	0.18	0.18	0.36	0	0	0.36
Mountain Whitefish	2010	50.6	0.88	0	0.72	0.12	38	0.4	9.1
	2011	96	0	0.15	0.18	2.1	0.68	0.51	0.51
	2017	75	0	5.0	0	4.9	9.5	0	5.2
	2018	76	0	1.0	0	0.19	20	0	2.8
Rainbow Trout	2010	52	36	0.53	0	10	0.18	0.029	0.74
	2011	93	0	0.03	0	5.3	0.4	0	1.7
	2017	1.8	0	81	0	14	0.16	0	3.3
	2018	5.4	0	69	0	15	1.9	0	8.8

Table 10: Relative abundance of prey items identified in the stomach contents of fish captured in the Peace River.

Species	Year	Relative Abundance (%)							
		True Flies	Mayflies	True Bugs	Leeches	Terrestrial	Caddis-flies	Copepods	Other Taxa
Arctic Grayling	2010	5.1	69	0.25	0	20	5.0	0	1.1
	2017	43	20	12	0.095	23	0.95	0	1.1
	2018	20	40	17	0.7	20	0.7	0	1.6
Kokanee	2010	9.8	60	0	0.75	19	0.75	0	9.8
	2011	12	9.0	0	0	4.8	0.11	73	1.1
Longnose Sucker	2011	100	0.19	0	0	0.07	0	0.023	0.16
Mountain Whitefish	2010	1.7	9.7	0	5.0	0.14	64	0	19
	2011	5.5	21	0	36	0.39	29	0	8.5
	2017	80	7.6	0.24	2.9	0.65	8.4	0	0.52
	2018	56	9.5	0	7.9	0.082	24	0	2.9
Rainbow Trout	2010	5.1	61	0	0	25	2.2	0.56	6.2
	2011	3.3	37	0.22	0	30	26	0	3.3
	2017	25	53	13	0.34	5.4	1.8	0	0.99
	2018	4.3	89	4.0	0.12	1.5	0.53	0	0.23

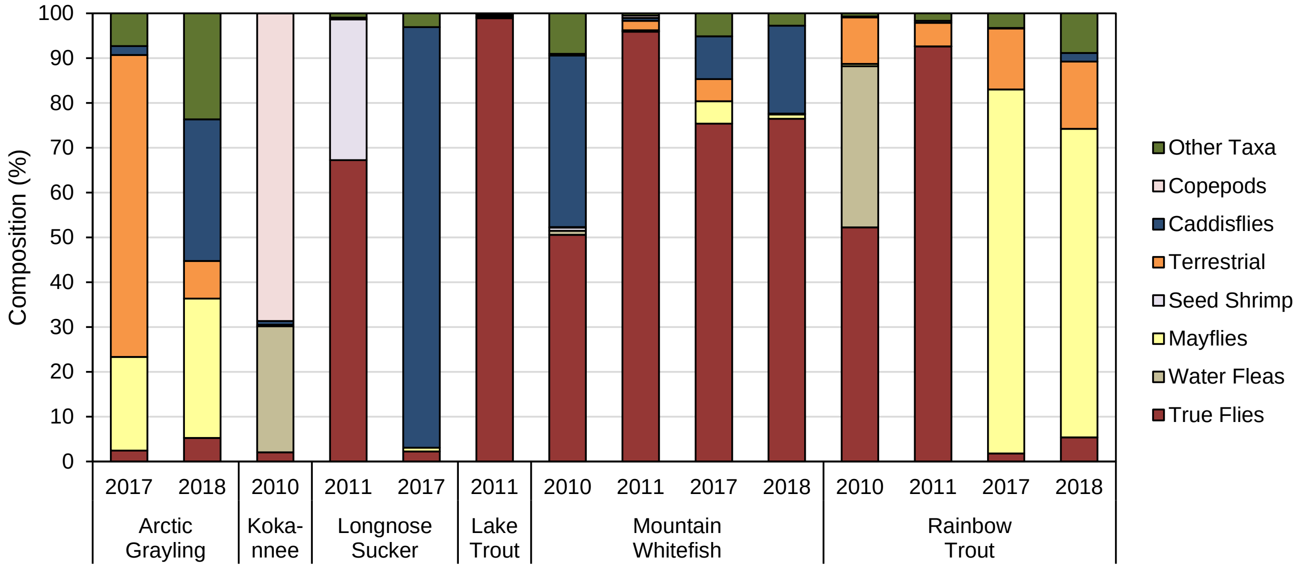

Figure 8: Relative abundance of prey items identified in the stomach contents of fish captured in Dinosaur Reservoir.

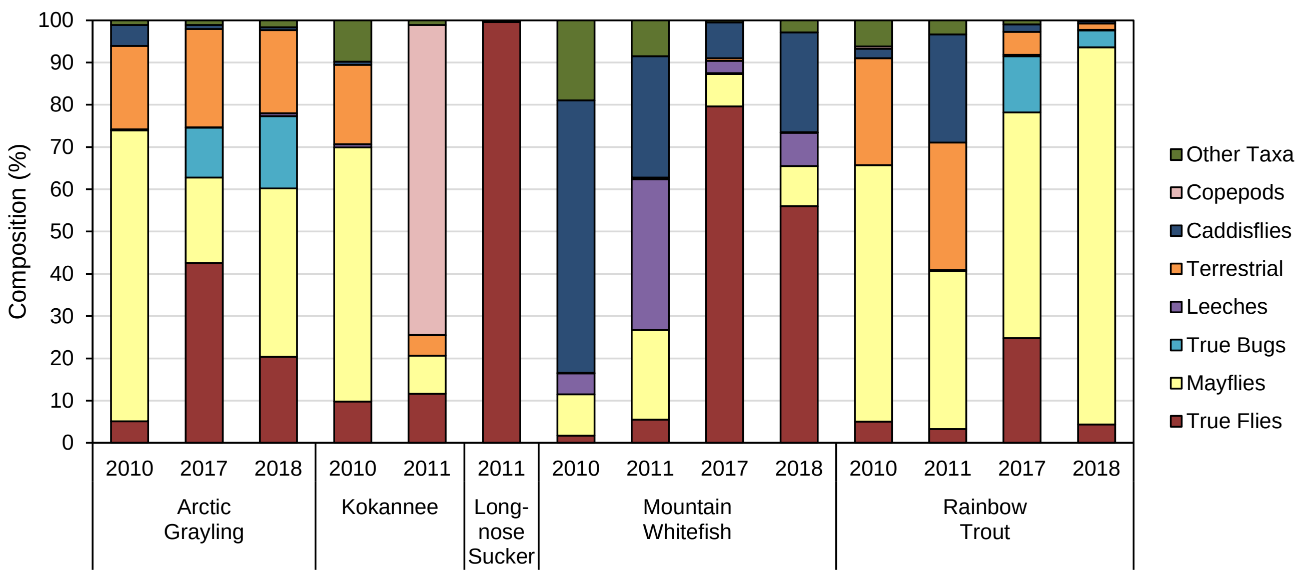

Figure 9: Relative abundance of prey items identified in the stomach contents of fish captured in the Peace River.

5.0 INTEGRATED FOOD WEB ANALYSIS

Table 11 and Table 12 summarize the likely prey items for sampled fish species in Dinosaur Reservoir and the Peace River based on an integration of the stable isotope analysis, stomach content analysis, and diet classification. Because fish were not recorded in stomach contents, the stomach content analysis was limited to invertebrates. Stable isotope data were limited to just three invertebrate taxa. There were some discrepancies between potential prey items identified by stable isotope analysis and likely prey based on diet classification as well as general knowledge of the aquatic ecosystem. This section includes a discussion of these discrepancies for each species.

Table 11: Summary of likely prey items of fish species sampled in Dinosaur Reservoir based on literature, $\delta^{15}N$, $\delta^{13}C$, and stomach content data.

Species	Diet Classification	Likely Prey Items			
		$\delta^{15}N$	$\delta^{13}C$	Stomach Contents[a]	Integrated[b]
Lake Trout	Piscivorous	BT, RB, MW, LSU	LT, BT, RB, MW, LSU, GA	DP	BT, RB, MW, LSU, DP
Bull Trout	Piscivorous	RB, MW, LSU	LT, BT, MW, LSU	-	MW, LSU
Rainbow Trout	Occasionally piscivorous	GA	LT, RB, MW, LSU, GA	DP, DS, EP.	DP, DS, GA, EP
Mountain Whitefish	Occasionally piscivorous	RB, MW, LSU, GA	LT, BT, RB, MW, LSU, GA	DP	DP, GA
Longnose Sucker	Non-piscivorous	LSU, GA	LT, BT, RB, MW, LSU, GA	-	GA

[a] Invertebrate species are identified as potential prey if the relative abundance in stomach contents was greater than 30%.
[b] Fish species are identified as potential prey if they were identified by both nitrogen and carbon stable isotope analysis, or by the stomach content analysis. Fish are removed from potential prey items if the consumer was identified as non-piscivorous in the literature review.
DP = Diptera; DS = diplostraca; GA = gastropods, EP = mayflies; TR = caddisflies, LT = Lake Trout, BT = Bull Trout; RB = Rainbow Trout; MW = Mountain Whitefish; LSU = Longnose Sucker.
"-" = data not available.

Table 12: Summary of likely prey items for fish species sampled in the Peace River based on literature, $\delta^{15}N$, $\delta^{13}C$, and stomach content data.

Species	Diet Classification	Likely Prey Items			
		$\delta^{15}N$	$\delta^{13}C$	Stomach Contents[a]	Integrated[b]
Lake Trout	Piscivorous	BT[c], BB, NP, GE, RB, MW, RSC, AG, LSU	LT, BB, NP, GE, RB, MW, LSU, GA, EP	-[d]	RB, MW, LSU
Walleye	Piscivorous	WP, BT, BB, NP, GE, RB, MW, RSC, AG, LSU	WP, BB, NP, GE, RB, MW, RSC, LSU, GA, EP	-	WP, BB, NP, GE, RB, MW, RSC, LSU
Bull Trout	Piscivorous	WP, BT, BB, NP, GE, RB, MW, RSC, AG, LSU, GA	BT, BB, NP, GE, RB, MW, AG, LSU, EP, TR	-	BT, BB, NP, GE, RB, MW, AG, LSU
Burbot	Piscivorous	BT, BB, GE, RB, MW, RSC, AG, LSU, GA, EP	LT, WP, BT, BB, NP, GE, RB, MW, RSC, AG, LSU, GA, EP, TR	-	BT, BB, GE, RB, MW, RSC, AG, LSU, GA, EP
Northern Pike	Piscivorous	BB, GE, MW, RSC, AG, LSU, GA	LT, WP, BT, BB, NP, GE, RB, MW, AG, LSU, GA, EP.	-	BB, GE, RB, MW, AG, LSU, GA, EP
Goldeye	Occasionally piscivorous	MW, RSC, LSU, GA, EP	LT, WP, BT, BB, NP, GE, RB, MW, RSC, LSU, GA, EP.	-	MW, RSC, LSU, GA, EP
Rainbow Trout	Occasionally piscivorous	MW, RSC, LSU, GA, EP	LT, WP, BT, BB, NP, GE, RB, MW, RSC, AG, LSU, GA, EP, TR	EP, TER	MW, RSC, LSU, EP, GA, TER
Mountain Whitefish	Occasionally piscivorous (small fish)	BT, BB, NP, GE, RB, MW, RSC, AG, LSU, GA, EP, TR	LT, WP, BT, BB, NP, GE, RB, MW, AG, LSU, GA, EP, TR	DP, HR, TR	GA, EP, TR
Redside Shiner	Occasionally piscivorous (small fish)	LSU, GA, EP.	WP, BB, GE, RB, RSC, LSU, GA, EP.	-	GA, EP.
Arctic Grayling	Occasionally piscivorous (small fish)	LSU, GA, EP.	BT, BB, NP, RB, MW, AG, LSU, EP.	DP, EP	DP, EP.
Longnose Sucker	Non-piscivorous	MW, LSU, GA, EP, TR.	LT, WP, BT, BB, NP, GE, RB, MW, RSC, AG, LSU, GA, EP, TR	DP	GA, DP, EP, TR.

[a] Invertebrate species are identified as potential prey if the relative abundance in stomach contents was greater than 30%.
[b] Fish species are identified as potential prey if they are identified by both nitrogen and carbon stable isotope analysis, or by the stomach content analysis. Fish are removed from potential prey items if the consumer was identified as non-piscivorous fish in the literature review.
[c] DP = diptera; DS = diplostraca; HR = leeches; GA = gastropods, EP = mayflies; TR = caddisflies, TER = terrestrial insects; LT = Lake Trout, WP = Walleye; BT = Bull Trout; BB = Burbot; NP = Northern Pike; GE = Goldeye; RB = Rainbow Trout; MW = Mountain Whitefish; RSC = Redside Shiner; AG = Arctic Grayling; LSU = Longnose Sucker.
[d] "-" = data not available.

5.1 Piscivorous Fish

Bull Trout

Bull Trout feeding habits vary with age. Juvenile Bull Trout tend to feed on aquatic invertebrates, but may also start feeding on fish within their first year (McPhail 2007). Adults tend to feed on trout, whitefish (especially Mountain Whitefish), Kokanee, Arctic Grayling, suckers, minnows, sculpins, and occasionally Redside Shiner (McPhail 2007). While adult Bull Trout may continue to feed on invertebrates, they are opportunistic in nature and may be entirely piscivorous given availability of prey (Wilhelm et al. 1999). Bull Trout were among the highest calculated trophic level in Dinosaur Reservoir and the Peace River, consistent with reported dietary piscivory (McPhail 2007). Comparing the one-source food web estimate, trophic position was higher in Dinosaur Reservoir (4.4) than the Peace River (3.8), which may indicate prey items but also may reflect the uncertainty in the estimation of the food base. Potential prey identified by stable isotope analysis were Mountain Whitefish and Longnose Sucker in Dinosaur Reservoir and Bull Trout, Northern Pike, Goldeye, Rainbow Trout, Mountain Whitefish, Arctic Grayling, and Longnose Sucker in the Peace River. No stomach content data were available for Bull Trout in either Dinosaur Reservoir or the Peace River. Kokanee are expected to be a common prey item for Bull Trout in Dinosaur Reservoir however stable isotope data were not available for Kokanee to confirm this hypothesis (McPhail 2007).

Northern Pike

Larval Northern Pike eat primarily aquatic invertebrates and become increasingly piscivorous as they grow. Northern Pike have been documented to seasonally alter the proportion of fish to invertebrates in their diet. Fish species preyed upon by Northern Pike include Longnose Sucker, Arctic Grayling, Mountain Whitefish, small Walleye, stickleback, chub, dace, and sculpins. Northern Pike were only collected from the Peace River, where potential prey identified by stable isotope analysis were Burbot, Goldeye, Rainbow Trout, Mountain Whitefish, Arctic Grayling, and Longnose Sucker. Stomach content data were not available for Northern Pike in either Dinosaur Reservoir or the Peace River.

Walleye

Larval Walleye are primarily planktivores but begin consuming fish at an early age, depending on the availability of prey (McPhail 2007). In the Peace River, Walleye likely become piscivorous by age-1. Fish species preyed upon by juvenile and adult Walleye include suckers, Mountain Whitefish, Burbot, Lake Chub (*Couesius plumbeus*),Yellow Perch (*Perca flavescens)*, Arctic Grayling, and other Walleye. Walleye are not present in Dinosaur Reservoir. Therefore, all collected samples were from the Peace River downstream of Peace Canyon Dam, where all fish species except Lake Trout and Bull Trout were identified as potential prey by stable isotope analysis. Stomach content data were not available for Walleye.

Burbot

Juvenile Burbot feed on benthic invertebrates (e.g., amphipods). Adult Burbot move into deeper water and increase the proportion of fish in their diet, which includes trout, Mountain Whitefish, Redside Shiner, Arctic Grayling, suckers, minnows, and sculpins (McPhail 2007, Baranowska and Robinson 2017). Burbot were only collected from the Peace River, where potential prey identified by stable isotope analysis included gastropods, mayflies, and all fish species present except Lake Trout, Walleye, and Redside Shiner. The possibility of cannibalism was not excluded by stable isotope analysis; cannibalism by Burbot is documented in other regulated rivers (Amundsen et al. 2003). Stomach content data were not available for Burbot in either Dinosaur Reservoir or the Peace River.

Lake Trout

Larval Lake Trout are planktivorous, switching to larger invertebrates (e.g., amphipods and molluscs) and fish as they grow larger. Adult Lake Trout may remain planktivorous if fish prey are not available. Adult Lake Trout may also switch to a planktivorous diet in the summer if constrained by a thermocline. In the current study, most Lake Trout were collected in the Dinosaur Reservoir; only three individuals were collected from the Peace River, and these likely were fish entrained from Dinosaur Reservoir. Lake Trout had the highest calculated trophic level in both Dinosaur Reservoir and the Peace River (along with Walleye), consistent with reported dietary piscivory (McPhail 2007). Potential prey identified by stable isotope analysis in Dinosaur Reservoir were Bull Trout, Rainbow Trout, Mountain Whitefish, and Longnose Sucker. These species, excluding Bull Trout, were also identified as potential prey in the Peace River along with Burbot, Northern Pike, Goldeye, and Longnose Sucker. However, Burbot, Northern Pike, and Goldeye are not expected overlap in habitat in the Peace River and are unlikely potential prey items (Mainstream 2012). Stomach content data for Lake Trout were limited to 26 samples collected in Dinosaur Reservoir in 2011. Information from these samples indicated that Lake Trout in Dinosaur Reservoir were feeding almost exclusively on true flies at the time of sampling.

5.2 Occasionally Piscivorous Fish

Goldeye

Adult Goldeye consume predominantly aquatic invertebrates, although larger individuals will consume small fish (McPhail 2007). Larval Goldeye feed on plankton and begin to consume insects at the fry stage. Goldeye are only present in the lower sections of the Peace River, where the estimated trophic position ranged from 3.5 to 3.9. This trophic position was moderate compared to other fish species, which is consistent with an occasionally piscivorous species. Potential prey identified by stable isotope analysis in the Peace River were Mountain Whitefish, Redside Shiner, Longnose Sucker, and gastropods. Other invertebrate prey items and vertebrate prey items (e.g., mice, voles that fall into the water) are expected to be potential dietary sources; however, data are not available to confirm this assumption. Stomach content data were not available for Goldeye.

Mountain Whitefish

Lacustrine Mountain Whitefish fry feed primarily on plankton and adults feed on plankton, snails, aquatic insects, and occasionally small fish (McPhail 2007). Fluvial population fry feed on small aquatic insects and adults feed on nymphs of aquatic insects and occasionally terrestrial insects. In the current study, Mountain Whitefish had moderate calculated trophic levels in Dinosaur Reservoir and the Peace River. Potential prey identified by stable isotope analysis in Dinosaur Reservoir were Rainbow Trout, Mountain Whitefish, and Longnose Sucker. Potential prey identified in the Peace River were Bull Trout, Northern Pike, Goldeye, Rainbow Trout, Mountain Whitefish, Arctic Grayling, Longnose Sucker, gastropods, mayflies, and caddisflies. However, given the small size of Mountain Whitefish, they likely consumed only the smallest of the above listed species (i.e., age-0 individuals). Further, Mountain Whitefish tend to reside in faster water whereas the smaller juveniles and fry of Goldeye, Bull Trout, and Northern Pike tend to reside in slower moving, backwater areas in streams and would likely not overlap in habitat. Based on the primarily invertebrate-based dietary habits reported by McPhail (2007), it is likely that invertebrates represent a large proportion of the Mountain Whitefish diet. Stomach content data for Mountain Whitefish sampled from Dinosaur Reservoir (2010, 2011, 2017, and 2018) indicate Mountain Whitefish diet is predominately composed of true flies and caddisflies. Mountain Whitefish sampled from the Peace River (2010, 2011, 2017, and 2018) consumed less true flies, on average, than Mountain Whitefish sampled from Dinosaur Reservoir but consumed more copepods, mayflies, and caddisflies.

Arctic Grayling

Adult Arctic Grayling feed on aquatic insects with larger adults occasionally feeding on small fish (McPhail 2007). Arctic Grayling were only collected from the Peace River where potential prey identified by stable isotope analysis were Longnose Sucker and mayflies. However, the trophic position calculated ranged from 3.1 to 3.6, which was consistent with the range in trophic positions calculated for a non-piscivorous fish (e.g., Longnose Sucker); therefore, Longnose Sucker are unlikely to be Arctic Grayling prey in the Peace River. Uncertainty in this assessment is high, as only four individuals were collected in this study. Stomach content data was available for two years of sampling of Arctic Grayling in Dinosaur Reservoir (2017 and 2018), where dietary items were dominated by terrestrial insects, mayflies and caddisflies. In the Peace River, stomach content data from 2017 and 2018 indicated that Arctic Grayling diets were predominately composed of true flies and mayflies.

Rainbow Trout

Fluvial Rainbow Trout fry, juveniles, and adults have a similar diet, consuming drifting and emerging stages of benthic, aquatic, and terrestrial insects (McPhail 2007). Lacustrine Rainbow Trout consume a more varied and broad diet than fluvial forms, including benthic amphipods, snails, and adult insects. Large individuals (>400 mm) in large lakes may become piscivorous. Rainbow Trout had moderate calculated trophic levels in Dinosaur Reservoir and the Peace River, consistent with an occasionally piscivorous species. Comparing the one-source food web estimates, trophic position was the same in both Dinosaur Reservoir and the Peace River (3.5). Potential prey items were not identified by stable isotope analysis in Dinosaur Reservoir, likely reflecting the limited sample of invertebrates collected (n = 4 gastropods). In the Peace River, potential prey items identified were Mountain Whitefish, Redside Shiner, Longnose Sucker, and gastropods. Stomach content data for Rainbow

Trout sampled in Dinosaur Reservoir (2010, 2011, 2017, and 2018) indicated that diets at the time of sampling were predominately composed of either mayflies or true flies. Stomach content data for Rainbow Trout sampled in the Peace River (2010, 2011, 2017, and 2018) indicate mayflies represented the predominate dietary item.

Redside Shiner

Redside Shiner prey items vary and include nymph, pupal, and adult aquatic and terrestrial insects, cladocerans, copepods, molluscs, and fish eggs and fry (McPhail 2007). Redside Shiner were only collected from the Peace River, where potential prey identified by stable isotope analysis were Longnose Sucker, gastropods, and mayflies. However, the trophic position ranged from 3.1 to 3.6, which is low compared to other fish species. This is consistent with the non-piscivorous dietary habits reported by McPhail (2007); therefore, it is unlikely that Longnose Sucker are prey items of Redside Shiner. Stomach content data were not available for Redside Shiner.

5.3 Non-Piscivorous Fish

Longnose Sucker

Lacustrine Longnose Sucker fry feed primarily on plankton whereas fluvial fry feed primarily on chironomids (McPhail 2007). Adult Longnose Sucker feed on benthic organisms, primarily insects including chironomid, caddisflies, and plecopteran (stonefly) larvae. Longnose Sucker had the lowest trophic position in the Peace River (range from 3.0 to 3.4) compared to other fish species, consistent with a non-piscivorous diet. The trophic position calculated in Dinosaur Reservoir, however, was moderate compared to the other fish species (3.7). This may reflect altered prey or altered $\delta^{15}N$ in prey in the Dinosaur Reservoir compared to the Peace River, although the results likely reflect the high uncertainty in the estimate of the base of the food web in Dinosaur Reservoir. Potential prey identified by stable isotope analysis were Longnose Sucker (i.e., carnivory) in both waterbodies, and additionally Redside Shiner, gastropods, mayflies, and caddisflies in the Peace River. Carnivory and piscivory are not consistent with dietary habits as reported by McPhail (2007), so it is likely that Longnose Sucker prey items were not collected in Dinosaur Reservoir. Stomach content data for Longnose Sucker sampled in Dinosaur Reservoir were available for two years (2011 and 2017) and results varied between years, with diets predominately composed of true flies (2011) or mayflies (2017). Stomach content data for Longnose Sucker sampled in the Peace River were available for 2011 and indicated Longnose Sucker diet at the time of sampling was almost exclusively composed of true flies.

5.4 Implications for Monitoring

As indicated on the EIS, altered abundances of different consumer groups of fish are expected as a result of the Project. It follows that changes to prey abundance and consumer feeding patterns may occur. Shifts in mean isotopic signatures in fish species may be used to monitor changes in the food web as the Project progresses. Shifts in the stable nitrogen isotope may suggest fish are shifting their food source and eating higher or lower on the food chain (i.e., changes in degree of piscivory and invertivory). Shifts in the carbon stable isotope may indicate a change the basal food source and may suggest fish are eating in different locations. Stomach content SIA data as well as invertebrate SIA data can help to define the isotopic signature of the basal food source and to identity whether isotopic shifts in fish tissue are due to a changing basal signature or changing foraging behaviour.

Different carbon signatures occur between littoral and pelagic areas, as well as fast- and slow-moving rivers. Shifts in mercury could indicate changes to amounts of mercury present, forms of mercury (e.g., greater methylmercury levels) or changes to dietary sources with different concentrations of mercury.

6.0 CLOSURE

This memo summarized existing stable isotope data, mercury data, and stomach content collected between 2010 and 2019 during the Project's Peace River Large River Fish Indexing Survey. Data were analyzed to describe general dynamics of the food web in Dinosaur Reservoir and the Peace River. These findings are intended to be used to identify potential changes to food web structure in the Peace River in response to the construction or operation of the Project.

We trust the above meets your present requirements. If you have any questions or comments, please contact the undersigned.

Golder Associates Ltd.

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Associate, Senior Environmental Scientist

KM/AdB/cmc

Attachments: Figure 1A: Sample Collection Locations

https://golderassociates.sharepoint.com/sites/107993/project files/6 deliverables/issued to the client_for wp/19121767-012-tm-rev1/19121767-012-tm-rev1-bch site c sia analysis and food web model 23apr_21.docx

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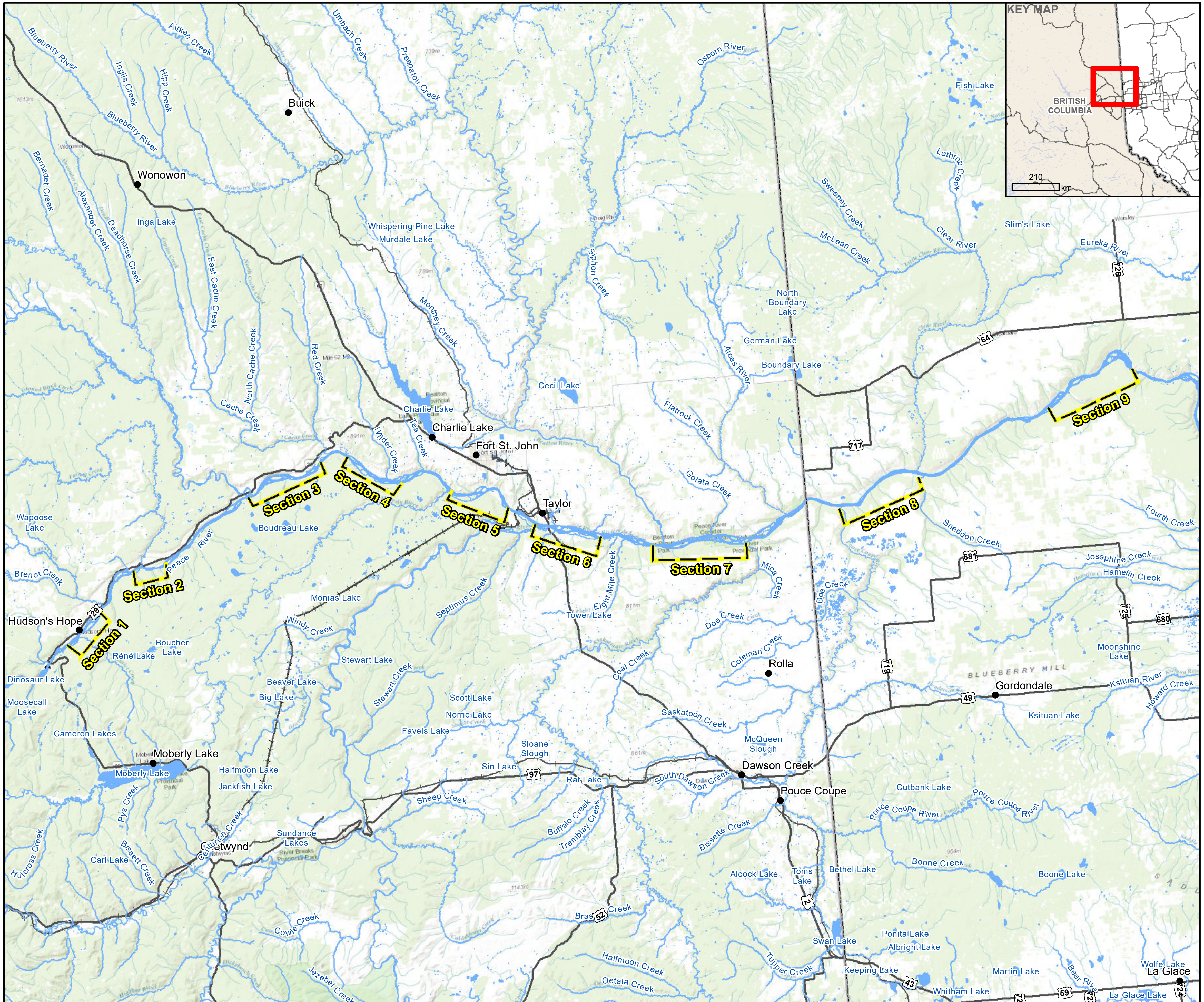

LEGEND

- PLACE NAME
- SITE C FAHMFP RIVER SECTIONS
- DAM SECTION
- PROVINCIAL BOUNDARY
- WATERCOURSE
- RAILWAY
- ROAD
- WATERBODY

0 10 20

1:625,000 METRES

REFERENCES
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2. DAM SITE OBTAINED FROM FROM GEOBASE®.
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CLIENT
BC HYDRO

PROJECT
SITE C FISH AND AQUATIC HABITAT MONITORING AND FOLLOW-UP PROGRAM

TITLE
PEACE RIVER FISH COMMUNITY MONITORING PROGRAM (MON-2)

CONSULTANT
GOLDER MEMBER OF WSP

YYYY-MM-DD	2020-11-20
DESIGNED	DF
PREPARED	CD
REVIEWED	KM
APPROVED	AdB

PROJECT NO.	PHASE	REV.	FIGURE
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IF THIS MEASUREMENT DOES NOT MATCH WHAT IS SHOWN, THE SHEET SIZE HAS BEEN MODIFIED FROM: ANSI B