

Fish Sampling at eDNA sites for Finlay Reach Arctic Grayling.



Photo: Ospika River mainstem

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Prepared By:

Mike Stamford, M.Sc.
Stamford Environmental
877 West Bay Road | Gambier Island, BC | V0N 1V0
michaelstamford430@gmail.com

Bryce O'Connor
ChuCho Environmental LLP
201-1116-6th Avenue
Prince George, BC.
V2L 3M6
bryce@chuchoenvironmental.com

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Table of Contents

Table of Contents	3
List of Figures	4
List of Tables	5
Executive Summary.....	6
1 Introduction	7
2. Goals and Objectives.....	11
3. Methods.....	12
3.1: Fish sampling	12
3.1.1: Beach Seine.....	13
3.1.2: Electrofishing	14
3.1.3 Angling.....	14
3.2 Environmental DNA sampling.....	14
4.0 Results and Discussion	15
4.1 Habitat Variables	15
4.2 Fish sampling effort	18
4.3 Fish species captured.....	19
4.4 2021 eDNA results	22
4.5 Reconciling the sparse distributions of Arctic Grayling in fish sampling, relative to eDNA detections.....	22
4.5.1 Ospika River	22
4.5.2 No Name Trib-4: Arctic Grayling eDNA detections downstream and upstream of impassable falls.	25
4.5.3 Aley Creek (eDNA site Trib-3).	26
4.5.4 Finlay Reach streams	26
5.0 Conclusions and recommendations	27
5.1 Conclusions:	27
5.2 Recommendations:.....	29
6.0 Literature.....	30

List of Figures

Figure 1: Arctic Grayling Critical habitats in Williston and Upper Peace core areas based on post 1988 fish collection records and observations. Purple=Adult (>200mm fork length); Green=Yearling (100-200mm fork length); Red=Young of the year (fry <100mm fork length). Figure adapted from Stamford et al. (2017).	8
Figure 2: Distribution of Arctic Grayling eDNA samples collected between 2018 and 2021 in Williston Reservoir (small pane) highlighting Finlay Reach streams (Large Pane): 1. Ospika River; 2. Lafferty Creek; 3. Collins Creek; 4. Davis River; 5. Chowika Creek; 6. Lower Finlay Core Area; 7. Ingenika Core Area. Arrows highlight Ospika tributaries sampled: i. Gauvreau Creek (Trib-1); ii. Stevenson Creek (Trib-2); iii. Aley Creek (Trib-3); iv. No Name Creek (Trib-4); v. Borden Creek (Trib-5); vi. McCusker Creek (Trib-6). <i>Green Dots</i> = Arctic Grayling eDNA detected; <i>Black Dots</i> = Arctic Grayling eDNA NOT Detected. Adapted from results in Stamford et al. (2020).	10
Figure 3: A typical site in Ospika River where beach seine hauls fished for Arctic Grayling fry and yearlings along the inside turn of the main channel. Beach seine shown in the foreground. Photo taken during sampling by Mike Stamford.	13
Figure 4: Meandering Lafferty Creek hypothesized to provide yearling habitat for Arctic Grayling and explain the downstream detection by eDNA. Both angling and electrofishing efforts covered one site within this reach. <i>Mike Stamford photo</i>	14
Figure 5: Distribution of August 2021 fish sampling sites (Black Dots) and locations of eDNA sites (Purple Arrows) collected from Finlay Reach streams. Stream lengths spanning the eDNA fish sampling are highlighted in purple. The pick and shovel icon shows the location of Aley Creek mine exploration site. Numbers labelling watersheds correspond with Figure 2: 1. Ospika River; i. Gavreau Creek (Trib-1); iii. Aley Creek (Trib-3); iv. No Name (Trib-4); 2. Lafferty Creek; 3. Collins Creek; 4. Davis River, 5. Chowika Creek. .	15
Figure 6: Average August daily discharge (Cubic Meters per Second, CMS) in Ospika River at Environment Canada's gauging station located upstream of Aley Creek (https://wateroffice.ec.gc.ca). The horizontal line highlights the average across 40 years at 37.5 CMS.	16
Figure 7: Gravel dominated stream margin and off-channel habitats observed upstream of eDNA site Ospika Main-3.	18
Figure 8: Species rare among fish captured in Ospika River watershed and adjacent Finlay Reach streams. A: Arctic Grayling fry (64mm) captured in reach OSPMain-2; B: Pygmy Whitefish (120mm) captured upstream at OSPMain-3; C: Longnose Sucker (158mm) captured upstream at OSPMain-4; D: Bull Trout (184mm) captured upstream of falls in Ospika Trib-4.	21
Figure 9: Distribution of Mountain Whitefish fry (green dots) captured in beach seines in Ospika River main channel. Arctic Grayling fry were captured at Fry-02. Blue curved arrows show confluences of Tributaries 'A' and 'B' relative to the fry captures (labelled in figure). Tributary A originates near the 'pick and shovel' icon that highlights the Aley Mine development.	23
Figure 10: The downstream reach of 'Tributary A' near the confluence with Ospika River that was never sampled for Arctic Grayling, neither with eDNA nor other fish sampling methods.	25
Figure 11: Sampling in unnamed Tributary 4 between 2019 and 2021. Labelled dots indicate fish and eDNA sample site locations (Green = Arctic Grayling eDNA detected; Black=	

Arctic Grayling eDNA not detected). Blue arrows indicate direction of stream flow, red slash indicate impassable (5meter high) falls.26

Figure 12: Length frequency distribution of Arctic Grayling fry captured during August 2017 in beach seine hauls in Finlay River (Upper Finlay Core Area). Fry were distributed along 50 km of main channel habitat. Coloured bars indicate three regions listed from upstream (Region A) to downstream (Region C). Data from Chu Cho 2017.28

List of Tables

Table 1: Fish sampling strategy proposed versus completed to evaluate Arctic Grayling life histories present upstream of eDNA sites where Arctic Grayling were detected in Ospika River watershed and four adjacent Finlay Reach streams in 2019. Six eDNA sites were resampled in 2021 to evaluate temporal consistency in their results, provide direct comparisons with fish sampling results, and confirm Arctic Grayling eDNA presence upstream of impassable falls. All sites were accessed using helicopter. 11

Table 2: Summary of habitat variable collected during fish sampling in Ospika watershed and adjacent streams draining the eastern slope of Finlay Reach. Finlay Reach streams are listed from south to north, and sites in Ospika watershed listed from downstream to upstream. 17

Table 3: Count of 2021 fish sampling events used to determine the Arctic Grayling life history stages present upstream of eDNA sites, which were collected in 2018 and 2019..... 19

Table 4: Detections among eDNA samples and field measurements of distances fished (in meters) among sampling events used to determine life history stages of Arctic Grayling present upstream of eDNA sites in Ospika and Finlay Reach watersheds. Cumulative distances fished (note the units change to kilometers) among eDNA sites assume and upstream detection distance of 1.5 km, and among fishing sites are the sum of field measurements. 19

Table 5: Number of fish and sizes captured while sampling in Ospika River and adjacent Finlay Reach streams (August 26 to 30, 2021). Tissue and fin ray samples were collected from Bull Trout, Rainbow Trout, and Arctic Grayling for genetic and age analyses.20

Table 6: Number of fish captured in streams sampled in Ospika and Finlay Reach watersheds.20

Table 7: Comparisons of August fish capture from similar reaches in electrofishing surveys historically (one site per stream; Langston and Blackman 1993) and most recently (in this 2021 survey) among the same Finlay Reach watersheds. Longer sample lengths in 2021 reflect multiple sites in Collins and Lafferty creeks and using sampling methods biased toward capturing Arctic Grayling. CCG= Slimy Sculpin; RB= Rainbow Trout; BT= Bull Trout; MW= Mountain Whitefish; LSU= Longnose Sucker..... 27

Executive Summary

This project addresses Research and Acquisition Priority 1 Action Number 9 in the FWCP Peace Region action plan by informing the distribution of critical habitats and begin identifying factors limiting Arctic Grayling population productivity (FWCP 2020). This conservation action is aimed to protect remnant populations of Arctic Grayling and was enabled by previous environmental DNA (eDNA) results, which informed recurring Data Gaps #7 and #8 in the FWCP Monitoring Framework. The detection of Arctic Grayling eDNA in five Finlay Reach streams expanded the known range of habitat use in Williston Core Area, a putative metapopulation assessed at high risk of extinction. This project completed inventory fish sampling upstream of 13 eDNA sites to address the hypothesis that Ospika River and adjacent small-to-medium size tributaries provide critical habitats for early life histories. Potentially, these streams draining the eastern slopes of Finlay Reach sustain an independent metapopulation (i.e., a new core area) and the eDNA results potentially signify the discovery of unique components of Arctic Grayling population diversity in Peace Basin.

Only two Arctic Grayling were captured in the whole survey. Among a total of 291 fish captured, 176 (60%) came from the main channel of Ospika River while the remaining 115 (40%) fish were captured from seven smaller streams, including three tributaries to Ospika River and four tributaries of Finlay Reach. Among seven fish species captured, six came from Ospika River main channel, and four species (Bull Trout, Mountain Whitefish, Rainbow Trout, Slimy Sculpin) were captured in tributaries. Pygmy Whitefish, Longnose Sucker, and Arctic Grayling were found only in Ospika River mainstem. Mountain Whitefish dominated the catch (72%) from Ospika River and Slimy Sculpin (53%) and Bull Trout (43%) dominated the catch from smaller streams. Beach seining targeted 11 low gradient sections (2.9 km cumulative sample length) of Ospika River main channel and captured 176 fish, including the two Arctic Grayling fry (60mm fork length) and 43 Mountain Whitefish fry (<70mm fork length). Fry of these two species often rear together in low gradient main channel habitats in other large rivers in Peace Basin.

The fish sampling result is the first to confirm that Arctic Grayling spawn and rear as fry (i.e., have natal areas) in Ospika River and supports the hypothesis that an independent metapopulation might reside in east Finlay Reach streams. Recurrent detections of Arctic Grayling eDNA upstream in Ospika River suggest that natal areas might be more widely distributed than suggested by fish sampling results. Given the importance of rare (low density) potentially unique locally adapted populations to species resilience, further sampling is recommended to investigate this hypothesis. Low density in Ospika River and absence of Arctic Grayling in fish sampling in adjacent Finlay Reach streams fails to support the hypothesis that eDNA detections of Arctic Grayling indicate increasing abundance. Although significant beach seining effort rigorously evaluated Arctic Grayling presence in Ospika River main channel, electrofishing and angling effort was thinly dispersed in off channel habitats and among smaller tributaries. Additional sampling effort is needed to determine Arctic Grayling life history stages associated with eDNA detections, determine the demographic structure, and conservation status of Finlay Reach Arctic Grayling.

1 Introduction

The Arctic Grayling (*Thymallus arcticus*) in the upper Peace Basin is of conservation concern and important to local First Nations, recreational anglers, and other community interest groups. Highly sensitive to land and water use practices that threaten freshwater habitats, the Arctic Grayling is a focal priority species for management by the Fish and Wildlife Compensation Program (FWCP) and British Columbia Ministry of Forests (MOF). Formation of Williston Reservoir flooded roughly 1,760 km² of Peace, Finlay, and Parsnip valleys, and resulted in the loss of 600 km of large river habitats. This resulted in serious declines in Arctic Grayling populations (Cannings and Ptolemy 1998; Sebastian et al. 2003) and substantially impacted, isolated, and displaced surrounding First Nations and other communities in upper Peace Basin (Baker et al. 2000; Littlefield et al. 2007). Flooding over 100 km of Finlay River seriously diminished habitats and ecological communities within Tsay Keh Dene First Nation territory and the Arctic Grayling is among those cherished species that suffered habitat loss and population declines due to flooding. This study is focused on streams draining the eastern slopes of Finlay Reach where recent environmental DNA (eDNA) sampling suggests that populations might be recovering or possibly adapting to flooded habitats. Metapopulations that survived flooding migrate almost exclusively among fluvial habitats, which suggests they might lack the necessary adaptations for adfluvial life history strategies and presence of the reservoir might restrict access to habitats (Clarke et al. 2007). Consequently, recovery of migratory behaviours that include movements through the Williston Reservoir might constitute ancestral genotypes most suited for survival in the newly flooded Peace Basin (Stamford et al 2022).

Among eight metapopulations (Core Areas) of Arctic Grayling identified in upper Peace Basin, those in Williston Core Area were assessed at High Risk of extinction due to rare accounts in fish sampling records (Figure 1; Stamford et al. 2015, 2017). Significant fish sampling efforts using multiple methods between 1988 and 1994 failed to capture or observe Arctic Grayling in streams draining the eastern slopes of Finlay Reach except for one location near the mouth of Chowika Creek (Williamson and Zimmerman 2004). This was in stark contrast to the mid 1970's when adult Arctic grayling in spawning condition were observed in five streams draining the eastern slopes of Finlay Reach, including Chowika Creek, Davis River, Collins Creek, Lafferty Creek, and Ospika River (Bruce and Starr 1985, but see Figure 22 in Stamford et al. 2017). The combined impacts from habitat loss, excessive exploitation, and changing conditions in the reservoir are thought to have contributed to a significant decline from previous records (Blackman 1992; Williamson and Zimmerman 2004). Genetic and microchemistry analyses among surviving metapopulations (core areas) suggest that those populations extirpated by flooding may have been distinct demographically and ecologically from surviving populations (Clarke et al. 2005; Stamford and Taylor 2005; Shrimpton and Clarke 2012). Potentially, flooding of Finlay River displaced once abundant large river adapted populations and subsequently their distribution was reduced from at least five streams to a single critical habitat where adults were captured near the flooded mouth of Chowika Creek (Figure 1).

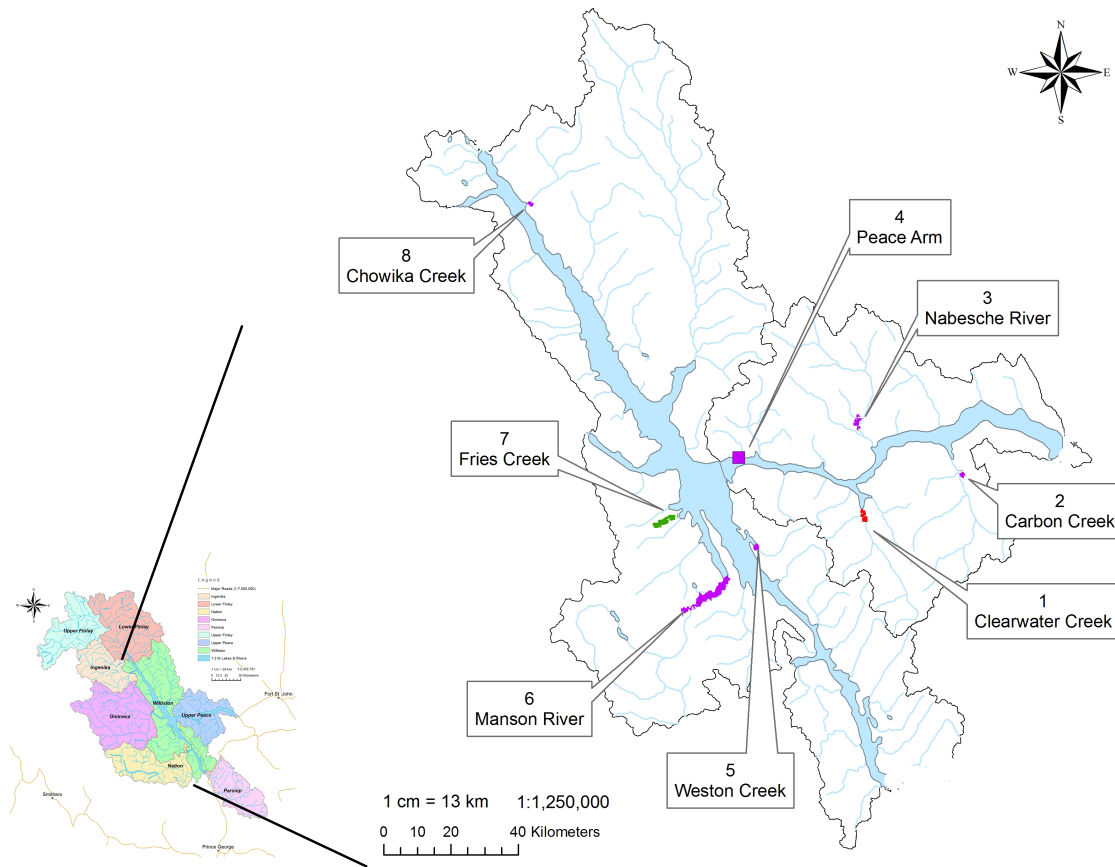


Figure 1: Arctic Grayling Critical habitats in Williston and Upper Peace core areas based on post 1988 fish collection records and observations. Purple=Adult (>200mm fork length); Green=Yearling (100-200mm fork length); Red=Young of the year (fry <100mm fork length). Figure adapted from Stamford et al. (2017).

More recently in 2018 and 2019, however, detection of Arctic Grayling eDNA found a wider range of habitat use, including all five streams draining the eastern slopes of Finlay Reach where adults were observed in the mid 1970's (Figure 2; Bruce and Star 1986). Possibly, the eDNA detections identify adult Arctic Grayling that migrated from natal areas situated in surrounding core areas (e.g., mainstems of Lower Finlay, and Ingenika core areas). Conceivably, populations might be recovering their ancestral migratory behaviours that now include movements through the reservoir. Strong site fidelity to summer rearing streams among adult Arctic Grayling might improve survival (e.g., Buzby and Deegan 2000) and individuals often migrate hundreds of kilometers through large rivers to rear with individuals originating from diverse natal streams (e.g., Blackman 2002; Earthtone and Mainstream 2013). Possibly, genetically based life history traits that promote homing to adult rearing locations in Finlay Reach streams survived flooding and metapopulations continue to migrate through Williston Reservoir as they did before large river habitats were enveloped by Williston Reservoir. Possibly, the recent eDNA results suggest a recovery of a migratory behaviour that is associated with multiple core areas.

Alternatively, the eDNA detections in these Finlay Reach streams have discovered rearing habitats for early life history stages (natal areas) and might signify a locally migrating metapopulation. A contiguous distribution of eDNA in the middle reaches of Ospika River (Figure 2) suggest that the associated low gradient areas in the main channel and downstream reaches of tributaries provide natal habitats for Arctic Grayling, similar to other core areas (e.g., Upper Finlay, ChuCho 2017; Ingenika, Lower Finlay). Furthermore, observations of spawning adults in the mid 1970's (Bruce and Starr 1985) and habitat modelling in Williston watershed (Hawkshaw et al. 2014) suggest that the sixth order Ospika River, and fifth order Chowika Creek and Davis River are sufficiently large to support all life history stages of Arctic Grayling. The eDNA detections in Davis River are in stark contrast to those in Ospika River, however, and suggest Arctic Grayling habitats are restricted to downstream reaches near the stream mouth (Figure 2). Possibly, physical habitat characteristics and fish species interactions limit the distribution of Arctic Grayling natal areas in Davis River and Chowika Creek, which both sustain large bull trout populations with extensive natal areas upstream of bedrock constrained rapids (Langston and Blackman 1993; Hagen et al. 2020).

Adjacent third order Collins and Lafferty Creeks might provide insufficient habitat areas and poor stability in water temperatures thought to limit natal habitats for Arctic Grayling in smaller streams (Hawkshaw et al. 2014). This suggests that multiple eDNA detections in sinuous low gradient reaches might provide rearing habitats for older life history stages, possibly including yearling sized (100-200mm) Arctic grayling (Figure 2). These types of habitats were seriously impacted by flooding and the loss is a plausible explanation for a population crash in Williston Core Area (see Stamford et al. 2017). Possibly, the detections of Arctic Grayling eDNA in Finlay Reach streams describe a metapopulation sustained by natal areas in Ospika River and a migratory behaviour that includes yearling and adult rearing habitats in Collins and Lafferty Creeks, and adult rearing in the lower reaches of Davis River and Chowika Creek.

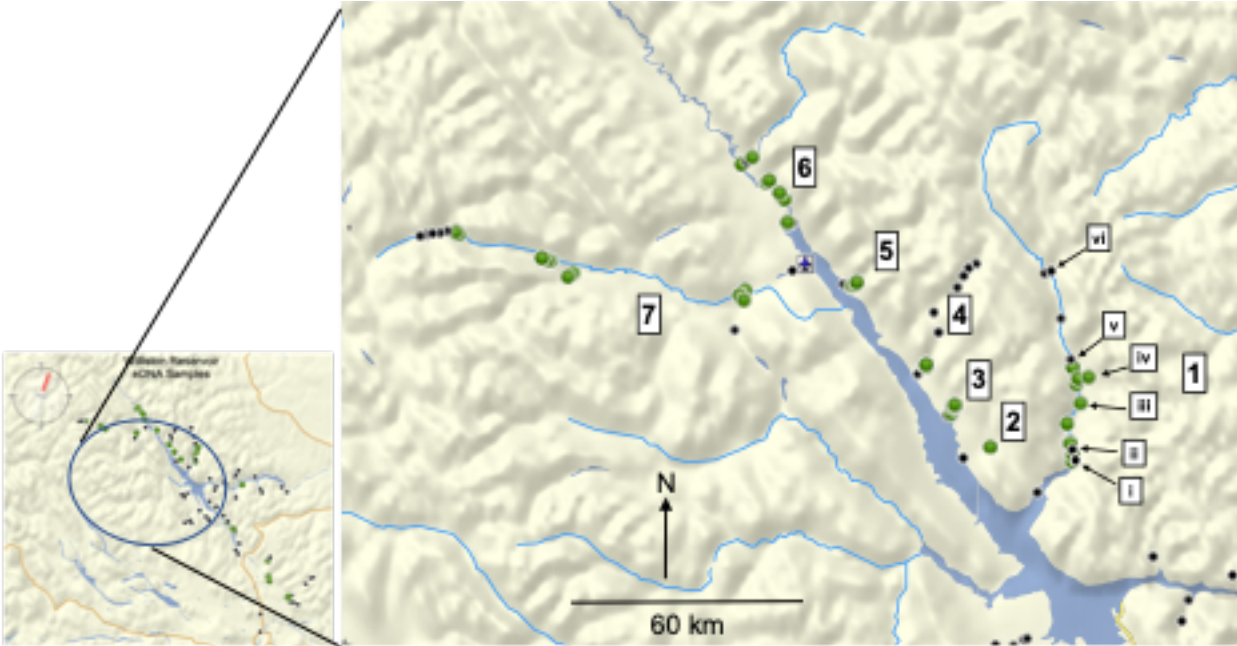


Figure 2: Distribution of Arctic Grayling eDNA samples collected between 2018 and 2021 in Williston Reservoir (small pane) highlighting Finlay Reach streams (Large Pane): 1. Ospika River; 2. Lafferty Creek; 3. Collins Creek; 4. Davis River; 5. Chowika Creek; 6. Lower Finlay Core Area; 7. Ingenika Core Area. Arrows highlight Ospika tributaries sampled: i. Gauvreau Creek (Trib-1); ii. Stevenson Creek (Trib-2); iii. Aley Creek (Trib-3); iv. No Name Creek (Trib-4); v. Borden Creek (Trib-5); vi. McCusker Creek (Trib-6). *Green Dots* = Arctic Grayling eDNA detected; *Black Dots* = Arctic Grayling eDNA NOT Detected. Adapted from results in Stamford et al. (2020).

Improved understanding of the critical habitats and factors that limit Arctic Grayling abundance in Finlay Reach streams will inform conservation and enhancement efforts needed to recover the historical abundance of Arctic Grayling in Williston Reservoir (Hagen and Stamford 2017). Possibly, this increased Arctic Grayling directed sampling efforts using highly efficient eDNA has identified a population that survived the flooding which, previous fish sampling efforts failed to detect (Langston and Blackman 1993; Williamson and Zimmerman 2004).¹

This project used fish sampling upstream of eDNA sites to distinguish between alternative hypotheses about the possible demographic structure in Finlay Reach streams (Figure 2). The first (null) hypothesis predicts that habitats upstream of eDNA sites support only adult Arctic Grayling: individuals large enough to migrate through the reservoir from distant natal areas. The second (alternative) hypothesis predicts that Ospika River and possibly the lower reaches of Chowika Creek and Davis River provide natal areas and fish sampling will identify rearing fry. Fish sampling in adjacent Collins and Lafferty creeks will hypothetically find only yearlings and adults. The results are anticipated to inform further studies aimed at understanding Arctic Grayling migratory behaviour, core area boundaries, and enable future conservation actions aimed to

¹ Fish sampling has significantly lower capture probabilities (~0.3 within the immediate sampled area) compared to eDNA sampling (~0.9 within 1.5km upstream; Wilcox et al. 2016; Stamford et al. 2022).

protect and define critical habitats and associated factors that limit population abundance in Williston Core Area.

2. Goals and Objectives

The project objective was to identify life history stages of Arctic Grayling associated with eDNA detections in five Finlay Reach streams and begin distinguishing between alternative hypotheses about possible demographic structure (Figure 2). Specifically, fish sampling methods most appropriate to capture young of the year (fry) and yearling life history stages were used to investigate the hypothesis that eDNA detections identify natal areas that potentially sustain an independent locally moving metapopulation (the alternative hypothesis). An ambitious fish sampling strategy (Table 1) had three priorities to direct efforts toward sample sites considered most likely to provide natal habitats for Arctic Grayling and to ensure informative data would be collected within the constraints of time and budget.

- Priority 1: Determine the presence of fry and juvenile life histories rearing in the extensive habitat areas associated with eDNA sites in Ospika River main channel and lower reaches of three tributaries.
- Priority 2: Determine the presence of fry and yearling life histories upstream of seven eDNA sites in four Finlay Reach streams adjacent to Ospika River.
- Priority 3: Determine the presence of adult life history stages in Finlay Reach streams.

Table 1: Fish sampling strategy proposed versus completed to evaluate Arctic Grayling life histories present upstream of eDNA sites where Arctic Grayling were detected in Ospika River watershed and four adjacent Finlay Reach streams in 2019. Six eDNA sites were resampled in 2021 to evaluate temporal consistency in their results, provide direct comparisons with fish sampling results, and confirm Arctic Grayling eDNA presence upstream of impassable falls. All sites were accessed using helicopter.

Site Location	No. eDNA sites evaluated 2019 *	No. Fish sampling sites proposed for 2021	Anticipated Method **	Fish Sample Sites Completed 2021	Method used 2021**
Ospika River main channel	4	12	SN, EF, AG, eDNA	20	SN, EF, AG, eDNA
Ospika River tributaries	3	12	SN, EF, AG, eDNA	15	EF, AG, eDNA
Finlay Reach streams	7	21	SN, EF, AG	8	EF, AG
Total	14	45		43	

* Arctic Grayling were detected at all eDNA sites except Gauvreau Creek, Ospika Tributary-1 .

** SN= Beach Seine; EF= Backpack electrofishing; AG= Angling

Following these priorities, significantly more fish sampling effort (150% more sites) evaluated the presence of Arctic Grayling natal areas in Ospika watershed, while sampling effort in Finlay Reach streams evaluated only 38% of sites planned for sampling (Table 1). Most of the sampling effort among Finlay Reach streams was in Collins and Lafferty creeks aimed to evaluate possible presence of juvenile (yearling) life history stages. Environmental DNA sampling was added to the sampling strategy to evaluate temporal consistency in detecting Arctic Grayling presence and collect data at the same time to compare with upstream fish sampling results in Ospika River

watershed main channel and two tributaries, including sites upstream of impassable falls in one tributary.

Results were anticipated to inform recurring data gap #7 (the unknown present-day distribution in Williston Core Area) and #8 (improve understanding of factors driving extirpation in small-to-medium size tributaries to the reservoir; Hagen and Stamford 2017). Addressing these data gaps will inform the conservation status and enable further conservation actions aimed to protect remnant populations in Williston Core Area. This project addresses Research and Acquisition Priority 1 Action Number 9 in the FWCP Peace Region action plan by informing the distribution of critical habitats and begin identifying factors limiting Arctic Grayling population productivity (FWCP 2020).

3. Methods

3.1: Fish sampling

Fish sampling took place over five days (August 26 through 30, 2021) using methods described in Johnson and Slaney (1996). Fish sampling at multiple sites upstream of each eDNA sites were selected in the field based on observations from the helicopter. Sampling effort in putative natal areas and yearling habitats in Ospika main channel was aimed at the low gradient braided reaches upstream of eDNA detections and sites were selected where the smaller substrate sizes indicated slow current along stream margins, usually located on the inside bends of the river. Arctic Grayling fry often accumulate in such habitats in large rivers (e.g. Finlay River; ChuCho 2017). The catch of Arctic Grayling fry from the Ospika main channel was used to select additional sites in the off-channel habitats (e.g. side channels, isolated pools, tributary confluences), assuming fry captured in the main channel originated from upstream spawning areas. Arctic Grayling spawning is often associated with tributaries and off channel habitats that provide productive fry rearing (e.g. Blackman and Hunter 2001) and fry densities often increase closer to spawning areas (Stuart and Chislett 1979; Butcher et al. 1981). Consequently, added sampling would hopefully help inform more precise locations of critical natal habitats.

Habitats selected in the smaller tributary streams were prioritized in the downstream reaches where Arctic Grayling fry often accumulate downstream from spawning areas. Selected sites included low-gradient areas with abundant cover (e.g. overhanging vegetation, woody debris, overhanging banks), slow riffles, shallow pools, and sinuous glides, which provide potential rearing habitats for Arctic Grayling fry and yearling life history stages.

Minimal time was spent searching for adult habitats in deeper pools and glides but these habitats were evaluated using angling with flies and lures if observed near landing sites or during flyover evaluations in the upstream reaches of the smaller streams.

Captured fish were processed at the end of each site and included species identification and fork-length for all species. Tissue clips preserved in 95% ethanol, and fin ray age structures preserved dried in envelopes were collected from Arctic Grayling, Bull Trout,

and Rainbow Trout for future studies (e.g. gene flow within and among core areas; age estimates).

Variables collected in the field for estimating sampling effort included stream length sampled, wetted widths within sampled lengths, electrofishing seconds. Distances and stream widths were measured using a range finder and GPS coordinates. Habitat variables were collected rapidly to roughly characterize sites, including wetted widths, water temperature, substrate composition, and D95 (Johnston and Slaney 1996). Stream lengths and gradients between eDNA sites and upstream fish sampling sites were estimated using GPS field coordinates and topographical data in Topo Canada V2 and the Garmin Base Camp (version 4.8.11, 2020).

3.1.1: Beach Seine

Stream margin habitats in Ospika River main channel were sampled using a 12-meter-long beach seine (3mm mesh). The net has proven effectiveness and appears to outperform electrofishing in capturing Arctic Grayling fry and yearlings from large river habitats (e.g. Omineca River, Schell et al. 2002; Kechika River, Degissi 2002; upper Finlay River, ChuCho 2017). Fish presence was evaluated using multiple beach seine hauls beginning at the upstream end of each site. The beach seine was hauled downstream with the current and captured fish rearing within the 12 m distance from the stream margin (Figure 3). Fish counts and seine haul lengths were recorded at the end of each haul and captured fish were carefully transferred into a bucket of river water. Seine hauls were repeated until increased flow rates pulled the leadline off the substrate. All observed habitats preferred by Arctic Grayling fry and yearlings were included in the beach seined site lengths.



Figure 3: A typical site in Ospika River where beach seine hauls fished for Arctic Grayling fry and yearlings along the inside turn of the main channel. Beach seine shown in the foreground. Photo taken during sampling by Mike Stamford.

3.1.2: Electrofishing

A Smith Root model LR 24 electrofisher was used to sample off-channel habitats in Ospika main-channel and low gradient riffle-pool type habitats in smaller streams. The unit settings were calibrated to local conditions then adjusted to minimize fish injury and optimize capture. Habitats considered preferred by Arctic Grayling fry and yearlings were high graded throughout a reach first in an upstream direction followed by a second pass downstream, which often improves the capture of Arctic Grayling. Other methods used to improve the capture of Arctic Grayling included ambushing certain habitats (e.g. undercut banks, pools), herding of fish from riffles into back channels.

3.1.3 Angling

Limited time was given to angling, which was done in pools and glides observed adjacent to eDNA, beach seine, and electrofishing sites. Two sites in upstream reaches in two streams (Aley Creek, Collins Creek) received only angling effort. Total time spent angling, total stream distance covered, and wetted width were used to measure sampling effort per angler at each site. Anglers used flies and lures and split up so each covered undisturbed adult habitats in the deeper pools and glides.



Figure 4: Meandering Lafferty Creek hypothesized to provide yearling habitat for Arctic Grayling and explain the downstream detection by eDNA. Both angling and electrofishing efforts covered one site within this reach. *Mike Stamford photo.*

3.2 Environmental DNA sampling

Environmental DNA was collected from six sites in Ospika watershed and took place before fish sampling or any other upstream activities (methods described in Stamford et al. 2022). The objectives for adding eDNA sampling in 2021 were:

1. To evaluate temporal consistency in detecting Arctic Grayling eDNA and provide real time comparisons with the fish sampling.
2. To confirm the 2019 results of Arctic Grayling eDNA detected upstream of impassable falls in Ospika Tributary-4.
3. Increase the number of trials comparing capture efficiencies between two different field methods (results presented in Stamford et al. 2022).

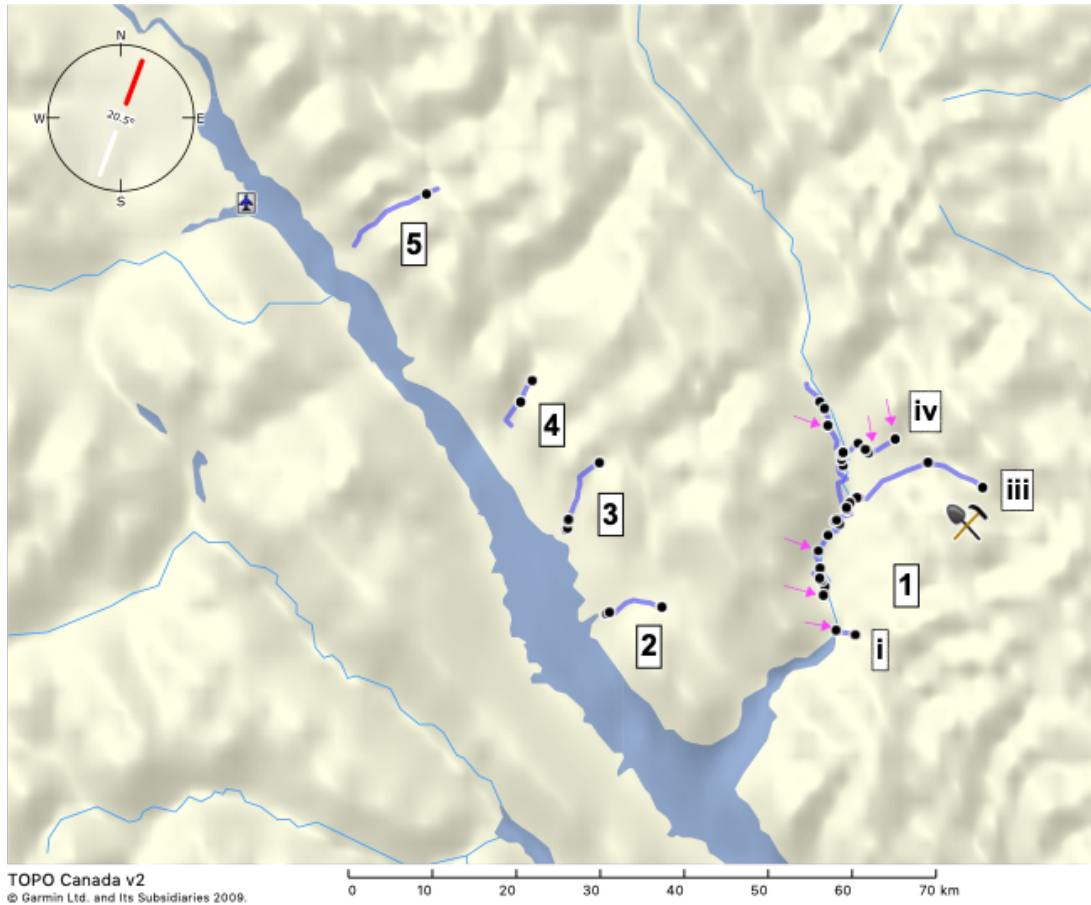


Figure 5: Distribution of August 2021 fish sampling sites (Black Dots) and locations of eDNA sites (Purple Arrows) collected from Finlay Reach streams. Stream lengths spanning the eDNA fish sampling are highlighted in purple. The pick and shovel icon shows the location of Aley Creek mine exploration site. Numbers labelling watersheds correspond with Figure 2: 1. Ospika River; i. Gavreau Creek (Trib-1); iii. Aley Creek (Trib-3); iv. No Name (Trib-4); 2. Lafferty Creek; 3. Collins Creek; 4. Davis River, 5. Chowika Creek.

4.0 Results and Discussion

4.1 Habitat Variables

About three days before sampling began the discharge in Ospika River dropped from 45 to 28 cubic meters per second (CMS) and then remained stable throughout the sampling period (August 26-30; <https://wateroffice.ec.gc.ca>). The fish, therefore, had time to recover from high water levels and establish in main channel rearing habitats before sampling began in Ospika River. Historically, the discharge during August in

Ospika River varied widely around the average. Possibly, the productivity of Arctic Grayling fry rearing areas might be limited in Ospika main channel by unpredictable discharge events. Attaining sufficient growth during their first summer is key to overwinter survival for Arctic Grayling and generally among temperate fish species (Northcote 1993).

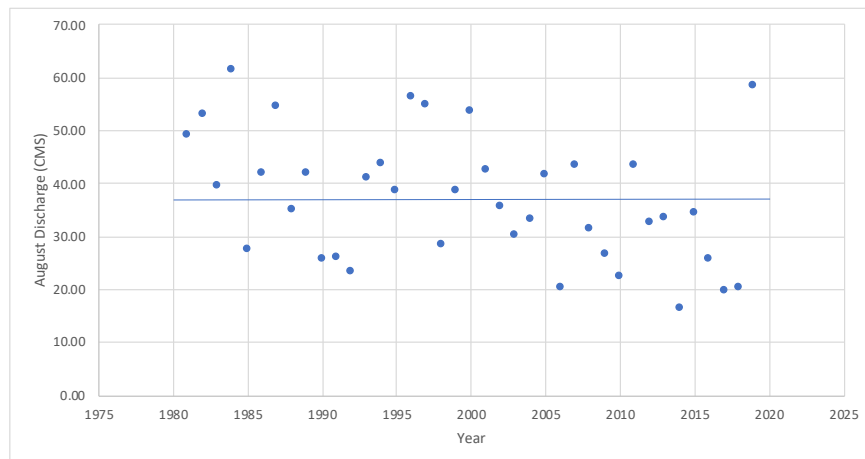


Figure 6: Average August daily discharge (Cubic Meters per Second, CMS) in Ospika River at Environment Canada's gauging station located upstream of Aley Creek (<https://wateroffice.ec.gc.ca>). The horizontal line highlights the average across 40 years at 37.5 CMS.

None of the streams were clear during sampling and turbidity in Ospika River was higher relative to the tributaries and Finlay Reach streams (Table 2). Water temperatures at fish sampling sites were generally colder than the preferred rearing temperature for Arctic Grayling adults (~10°C) and especially fry (16.6°C) as estimated in other Williston Core Areas (Hawkshaw et al. 2014). Recordings of temperature from other sources suggest, however, that August temperatures are often warmer in Finlay Reach streams. For instance, Langston and Blackman (1993) recorded a similar August temperature near our sample site in Chowika Creek (8°C) but Davis River (11°C), Lafferty Creek (11°C), and Collins Creek (11°C) were all warmer. Before our sampling period began, water temperature in Ospika River rose above 10°C and remained high (>11 °C) until about August 26 when the temperature dropped below 9 °C for the remainder of the summer (<https://wateroffice.ec.gc.ca>). The recent flood event appears to have influenced the temperature in Ospika main channel and might explain the relatively cold temperatures measured at fish sample sites (Table 2). Potentially, temporally variable temperatures recorded in Finlay Reach streams suggest warm ground water or tributary sources have greater influences during lower discharge and Arctic Grayling fry rearing habitat areas vary accordingly. Interestingly, our single recording of 12.8°C came from an electrofishing site at a tributary confluence near site Main-1 (Table 2).

Substrates at fish sample sites were observed to be mostly small, dominated by gravel and cobble sized D95 (Table 2), which indicates they were influenced by slower flow rates. Substrate size was an important criterion used to select sample sites in putative

Arctic Grayling fry rearing areas and an increasing D95 further upstream in Ospika River (i.e., sites Main-3 and Main-4; Table 2) reflect a more confined channel and difficulty in identifying at least three sample sites. Off channel habitats received minimal sampling effort due to budgetary constraints and included only three sites in downstream reaches where Arctic Grayling were encountered. Upstream reaches received zero effort in the off-channel habitat even though an abundance of habitat was observed (e.g., Figure 7).

Table 2: Summary of habitat variable collected during fish sampling in Ospika watershed and adjacent streams draining the eastern slope of Finlay Reach. Finlay Reach streams are listed from south to north, and sites in Ospika watershed listed from downstream to upstream.

Watershed	Stream Number and Name*	Water temperature (oC)	Turbidity**	Wetted Width (m)	D95 (cm)	Substrate Composition***
Finlay Reach	2. Lafferty	8.9	L	7.6	6.8	GFC
	3. Collins	8.7	L	6.8	18.8	CGF
	4. Davis	8.9	L	15.0	10.0	GFC
	5. Chowika	8.9	L	15.0	15.0	GFC
Ospika Tributaries	i. Gauvreau Creek (Trib-1)	8.0	L	15.0	20.0	CGF
	iii. Aley Creek (Trib-3)	8.0	L	10.3	14.5	CGB
	iv. No Name (Trib-4)	7.2	M	7.3	15.0	CGB
Ospika River	1. OspikaMain-1	8.0-12.8	M	64.1	15.0	GFC
	1. OspikaMain-2	8.3	M	60.4	15.0	GCF
	1. OspikaMain-3	8.0	M	36.6	18.0	GCF
	1. OspikaMain-4	9.2	M	35.0	22.0	GCF

* Stream numbers correspond to Figure 5.

** T=Turbid; M=Moderately Turbid; L= Lightly Turbid; C= Clear.

*** Listed in order from Dominant to Subdominant bed material: R=Rock (>400cm); B= Boulder (>25.6cm); C=Cobble (6.4-25.6cm); G=Gravel (0.2-6.4cm); Fines (<0.2cm).



Figure 7: Gravel dominated stream margin and off-channel habitats observed upstream of eDNA site Ospika Main-3.

4.2 Fish sampling effort

Among a total of 43 fish sampling events used to evaluate fish presence upstream of the eDNA sites, 81% were in Ospika watershed, and among those 57% were in the main channel. The eDNA sites in the downstream reaches of Ospika River (Main-1 and Main-2) received more fish sampling intensity, including both beach seining and electrofishing for two reasons: 1. More low gradient habitat areas were identified, relative to upstream; 2. Collections of Arctic Grayling fry directed added sampling in the off-channel habitats upstream of Main-1 and Main-2. Only eight sample sites (19%) evaluated the life histories present in the four Finlay Reach streams adjacent to Ospika River, including only a single electrofishing site upstream of each eDNA site in Davis River and Chowika Creek (Table 2; Figure 5). The beach seine and the electrofisher were used most frequently among sampling methods (67% of sites) while angling and eDNA sampling was used at 19% and 14% of sites, respectively (Table 3).

Table 3: Count of 2021 fish sampling events used to determine the Arctic Grayling life history stages present upstream of eDNA sites, which were collected in 2018 and 2019.

Watershed	eDNA (2019) Sample Site Location	eDNA site coordinates	Number of upstream Fish Sampling Sites by Method				Total Sampling events
			Angling	Beach Seine	Electrofishing	eDNA (2021)	
Finlay Reach	Lafferty Creek	10 V 423174 6247362	1		2		3
	Collins Creek-mouth	10 V 414843 6256032			1		1
	Collins Creek upstream	10 V 415522 6258506	1		1		2
	Davis River	10 V 409613 6269162			1		1
	Chowika Creek	10 V 395107 6290711			1		1
Ospika Tributaries	Ospika_Gauvreau Creek	10 V 441473 6243766	2		2	1	5
	Ospika_Aley Creek	10 V 442841 6258319	2		3		5
	Ospika_Trib-4-mouth	10 V 442543 6264742			1		1
	Ospika_Trib-4-upstream of falls	10 V 444679 6265197	1		1	2	4
Ospika Main Channel	OspikaMain-1	10 V 440509 6247947	1	3	1	1	6
	OspikaMain-2	10 V 439837 6253061		4	2	1	7
	OspikaMain-3	10 V 442129 6263227		3			3
	OspikaMain-4	10 V 441378 6267709		3		1	4
Total Sample Sites		13	8	13	16	6	43

Table 4: Detections among eDNA samples and field measurements of distances fished (in meters) among sampling events used to determine life history stages of Arctic Grayling present upstream of eDNA sites in Ospika and Finlay Reach watersheds. Cumulative distances fished (note the units change to kilometers) among eDNA sites assume upstream detection distance of 1.5 km, and among fishing sites are the sum of field measurements.

Watershed	eDNA Sites	*Arctic Grayling eDNA Detections		Stream distance sampled (m)			Total Distance sampled (m)
		2018/19	2021	Angling	Beach Seine	Electrofishing	
Finlay Reach	Lafferty Creek	1		620		622	1242
	Collins Creek	11		300		650	950
	Davis River	1				450	450
	Chowika Creek	1				300	300
Ospika Tributaries	Trib-1 (Gauvreau Creek)	0	0	496		296	792
	Trib-3 (Aley Creek)	1		1850		364	2214
	Trib-4 (No Name Cr)	11	00	1000		230	1230
Ospika Main Channel	Ospika R. Main-1	1	1	40	766	500	1306
	Ospika R. Main-2	1	1		1049	600	1649
	Ospika R. Main-3	1			757		757
	Ospika R. Main-4	1	1		380		380
Cumulative Distance Fished (km)		19.5	9	4.3	3.0	4.0	11.3

* each symbol represents a sample site; 1=GR eDNA detected; 0=GR eDNA Not detected

A total of 291 fish were captured from 11.3 km of sampled stream habitat, most of which (70%) was in the Ospika River and captured only two Arctic Grayling fry at a single site (Table 4; Figure 5). In contrast, the 13 eDNA sites sampled in 2018 and 2019 evaluated 73% more stream distance than all other fish capture methods combined, and all except one site detected Arctic Grayling eDNA (Table 4). Among the three other fish sampling methods, angling sampled across the longest stream distance and captured the fewest fish (Table 4, 5). Beach seining, however, sampled along the fewest kilometers of stream habitat but captured the most fish in numbers and diversity relative to other fishing methods (Table 4, 5). Beach seining captured 56.2 fish per kilometer sampled in Ospika River while electrofishing captured 30.2 fish per km from the smaller streams.

4.3 Fish species captured

Seven fish species were captured in total, and the Mountain Whitefish (*Prosopium williamsoni*) was by far the most numerous, accounting for 80% of fish captured in Ospika River and 55% overall (Table 5). Pygmy Whitefish (*Prosopium coulteri*), Arctic

Grayling, and Longnose Sucker (*Catostomus catostomus*) were rare in the samples and were captured only in Ospika River (Table 6; Figure 8). The two Arctic Grayling fry were captured in the same seine haul located about 500 m upstream from eDNA site Main-1 (Table 6; Figure 9). Bull trout (*Salvelinus confluentus*) and Slimy Sculpin (*Cottus cognatus*) accounted for 43% and 53%, respectively, of fish captured from the tributaries entering Ospika River and Finlay Reach streams (Table 6). Two Rainbow trout and two Mountain Whitefish were the only other fish species captured outside of Ospika main channel (Table 6).

Table 5: Number of fish and sizes captured while sampling in Ospika River and adjacent Finlay Reach streams (August 26 to 30, 2021). Tissue and fin ray samples were collected from Bull Trout, Rainbow Trout, and Arctic Grayling for genetic and age analyses.

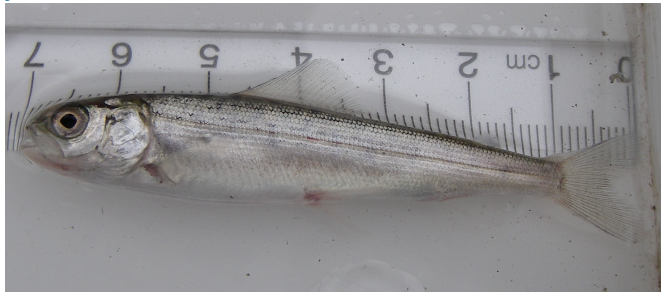
Fish Species	No. Fish Captured			Average FL (range, mm)
	Angling	Beach Seine	Electrofishing	
Arctic Grayling, <i>Thymallus arcticus</i>		2		62.0 (60-64)
Bull Trout, <i>Salvelinus confluentus</i>	3	8	53	145.6 (41-750)
Longnose sucker, <i>Catostomus catostomus</i>		3	2	122.6 (65-190)
Mountain whitefish, <i>Prospium williamsoni</i>		125	3	96.0 (29-248)
Pygmy Whitefish, <i>P. coulteri</i>		1		120.0
Rainbow Trout, <i>Oncorhynchus mykiss</i>	1		1	248.5 (122-375)
Slimy Sculpin, <i>Cottus cognatus</i>		27	62	47.4 (20-101)
Total	4	166	121	

The majority (60%) of all fish were captured in Ospika River main channel and among those 94% (166 fish) were captured using the beach seine, 4% (seven fish) were captured by electrofishing, and 2% (three fish) were captured angling (Table 5, 6). Among the 40% (115) of fish captured in tributaries and Finlay Reach streams all except one fish were captured electrofishing (Table 6). The four fish captured angling included three bull trout from Ospika River and one Rainbow Trout (*Oncorhynchus mykiss*) captured in Gauvreau Creek (Table 6).

Table 6: Number of fish captured in streams sampled in Ospika and Finlay Reach watersheds.

Fish species captured	Ospika Watershed				Finlay Reach streams				Total
	Ospika River	Gauvreau Creek (Trib-1)	Aley Creek (Trib-3)	No Name (Trib-4)	Lafferty Creek	Collins Creek	Davis River	Chowika Creek	
Arctic Grayling	2								2
Bull Trout	14	9	12	7	3	7	2	10	64
Longnose sucker	5								5
Mountain whitefish	126						2		128
Pygmy Whitefish	1								1
Rainbow Trout		1			1				2
Slimy Sculpin	28	3	3	4	28	23			89
Total	176	13	15	11	32	30	4	10	291

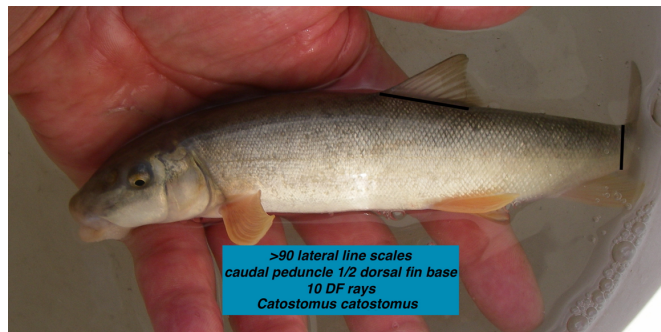
A: Arctic Grayling *Thymallus arcticus*



B: Pygmy Whitefish *Prosopium coulteri*



C: Longnose Sucker *Catostomus catostomus*



D: Bull Trout *Salvelinus confluentus*



Figure 8: Species rare among fish captured in Ospika River watershed and adjacent Finlay Reach streams. A: Arctic Grayling fry (64mm) captured in reach OSPMain-2; B: Pygmy Whitefish (120mm) captured upstream at OSPMain-3; C: Longnose Sucker (158mm) captured upstream at OSPMain-4; D: Bull Trout (184mm) captured upstream of falls in Ospika Trib-4.

4.4 2021 eDNA results

The 2021 eDNA results were consistent with results from previous years (2018/19; Stamford et al. 2020) at four out of five sites, including Arctic Grayling eDNA not detected in Gauvreau Creek, and detected at four sites in Ospika River (Main-1, Main-2, and Main-4; Table 4; Figure 5). Arctic Grayling were detected by both eDNA and fish collection methods at only one site in Ospika River main channel, located upstream of site Main-1. The 2021 eDNA sampling upstream in Trib-4, however, failed to detect Arctic Grayling at two sites and suggests the 2019 detection of Arctic Grayling eDNA upstream of impassable falls was an error (see section 4.5.2 for further discussion).

4.5 Reconciling the sparse distributions of Arctic Grayling in fish sampling, relative to eDNA detections.

4.5.1 Ospika River

The catch of Arctic Grayling (0.6 per km) and Mountain Whitefish fry (42 per km) in beach seine samples suggests that Ospika River is limiting in fry rearing habitats relative to large rivers in other core areas. For instance, using the same beach seine to sample in similar low gradient habitats in Finlay River found 141 Arctic Grayling (70 GR/km) and 338 Mountain Whitefish (169 MW/km) fry (ChuCho 2017). Nonetheless, by capturing two Arctic Grayling fry the results confirm the presence of natal habitats in Ospika River main channel, and consistent detections of Arctic Grayling eDNA both in 2019 and 2021 including three Ospika main channel sites (Table 4) suggests that natal habitats might be more widely distributed than the fishing results suggest. Possibly, rare Arctic Grayling fry escaped capture in the less efficient fish sampling (Table 2). Possibly, the distribution of Mountain Whitefish fry might help inform habitat use by the Arctic Grayling fry in Ospika River and explain eDNA detections (Figure 3).

For instance, rearing habitats for fry and yearling often overlap between Arctic Grayling and Mountain Whitefish in large river habitats where the current is slowest along stream margins and in off-channel habitats (reviewed in Stamford et al. 2017). As the current increases, however, only Mountain Whitefish continue to be present while the Arctic Grayling tend to drop out of beach seine captures (ChuCho 2017). Possibly, slow current areas preferred by Arctic Grayling are limiting in the low gradient braided areas in Ospika River and our beach seining effort failed to find them except at one site. Potentially, the patchy distribution of the more abundant Mountain Whitefish fry (FL<80mm) in our samples might suggest low densities of Arctic Grayling fry also rear in the same areas (Figure 9). Arctic Grayling fry might frequent the off-channel habitats which received minimal sampling efforts in 2021. Alternatively, eDNA sampling might have detected presence of adult or subadult Arctic Grayling rearing in Ospika mainstem. Although larger adults find forage habitats further upstream near headwaters in watersheds, smaller subadults and adult often rear in glides and pools distributed downstream, including mainstem habitats (e.g., Chowika Creek, Figure 1; Tack 1980; Hughes 1999; Baccante 2010). Increased sampling effort in the main channel and off-channel habitats directed at the locations where whitefish fry were captured (Figure 9) will help inform the contrasting successes between eDNA and fish sampling in detecting Arctic Grayling presence in Ospika River

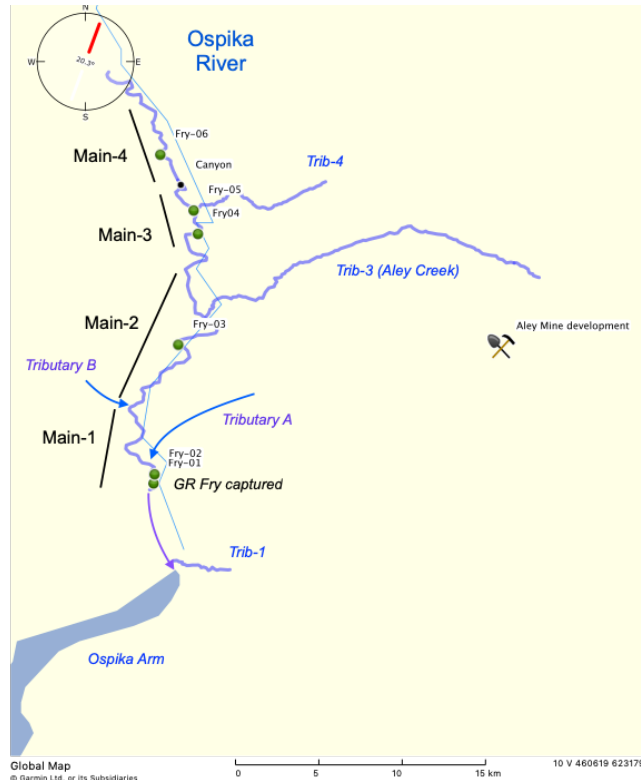


Figure 9: Distribution of Mountain Whitefish fry (green dots) captured in beach seines in Ospika River main channel. Arctic Grayling fry were captured at Fry-02. Blue curved arrows show confluences of Tributaries 'A' and 'B' relative to the fry captures (labelled in figure). Tributary A originates near the 'pick and shovel' icon that highlights the Aley Mine development.

Arctic Grayling fry show a strong preference for warm (~16°C) rearing habitats (e.g., Parsnip and Nation core areas; Hawkshaw et al. 2014). Warm water temperatures in natal areas promotes rapid growth rates, diminishes predation from cold adapted fish species (e.g., char), which together improves survival in Arctic Grayling (Northcote 1993). Selection pressures promote fine tuning of the life history traits associated with their growth response to natal temperatures, where locally adapted fry grow slower in natal habitats colder and warmer than their home streams (Haugen and Vøllestad 2000). Possibly the cold temperatures measured in Ospika watershed (Table 2) suggests warm natal areas are limiting for Arctic Grayling and the captures of only two fry (Table 5) indicate marginal habitats. Possibly, Ospika River is unable to sustain a metapopulation and the fry originated from adults that strayed from other core areas. Alternatively, the fry originated from adults that home to spawning sites where warm ground water sources and tributaries provide critical natal habitats for fry. Possibly, interactions with other fish species (e.g., Bull Trout predation, competition with Kokanee) has promoted further specialization and constrained Arctic Grayling to natal habitats that provide an escape from predation and competition. Possibly, recent increased abundance of introduced Columbia origin Kokanee in Ospika River have also increased competition to the detriment of Arctic Grayling (Wilson and Shrimpton 2020)

Interestingly, sympatric Arctic Grayling and Bull Trout in upper Omineca River appear to sort their natal streams according to temperature among adjacent tributaries (Schell 2002). Arctic Grayling fry and juveniles predominated in two warm (~16°C) tributaries while Bull Trout fry and juveniles were found exclusively in adjacent cold (~8°C) tributaries. Possibly, the Arctic Grayling fry captured in beach seines in Ospika River originated from warm habitat patches and further sampling in tributaries or associated off channel habitats will find higher fry abundance and improve descriptions of critical habitats and inform limiting factors associated with this apparently small population.

Two Ospika tributaries never received eDNA sampling and remain unsampled², for instance, and their confluences are just upstream from where the Arctic Grayling fry were captured (Tributary A, Tributary B; Figure 9). The downstream reaches of both tributaries drain extensive low gradient areas near the confluence with Ospika River where habitat characteristics appear to provide stable and possibly warm natal areas (Figure 10). Electrofishing the off-channel habitats near the confluences with these tributaries (sites OSPMain-BC02 and BC03) and previous environmental assessments in Tributary A (Aley Corporation 2014) have failed to find Arctic Grayling. Nonetheless, 12.8°C was measured while sampling the off channels near Tributary B, which suggests further investigations might find rearing areas for Arctic Grayling fry. Given the importance of temperature on survival and local adaptation in Arctic Grayling natal areas, warm tributaries and groundwater sources such as this require future investigations aimed at finding critical habitats for Arctic Grayling in Ospika River.

² Stevenson Creek also remains unsampled for fish because eDNA sampling failed to detect Arctic Grayling. Previous sampling, however, found Bull Trout, Rainbow Trout, and Slimy Sculpin (Mark Shrimpton, pers. com.).

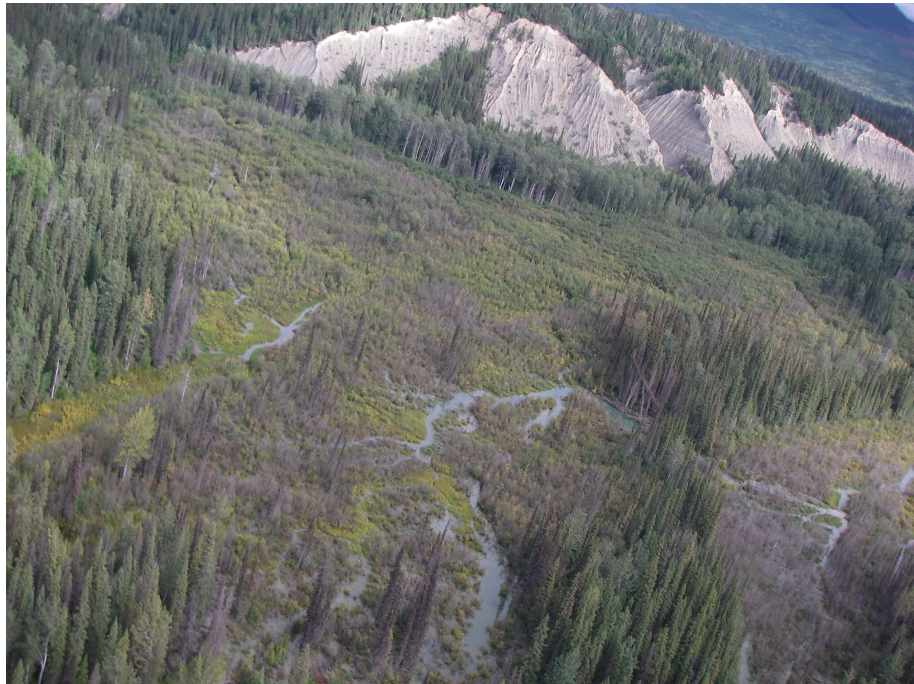


Figure 10: The downstream reach of 'Tributary A' near the confluence with Ospika River that was never sampled for Arctic Grayling, neither with eDNA nor other fish sampling methods.

4.5.2 No Name Trib-4: Arctic Grayling eDNA detections downstream and upstream of impassable falls.

Fish sampling in Tributary 4 was addressing the hypothesis that eDNA detected Arctic Grayling living both upstream and downstream of impassable falls. The upstream detection was hypothesized to signify headwater exchange with the upper Graeme River, one of the few streams where Arctic Grayling inhabit the headwaters (e.g., Lady Laurier Lake). Two electrofishing sites, one angling site, and two eDNA sites all failed to detect Arctic Grayling in Tributary-4 during August 2021 (Figure 11). The eDNA sampling in 2021 failed to detect Arctic Grayling using digital droplet PCR (ddPCR) at two sites upstream of the falls, which prompted a closer look at the original 2019 qPCR results. In 2019 the upstream sample yielded an "uncertain positive, at the limits of the assay detection", while the sample downstream of the falls yielded a firm detection of Arctic Grayling eDNA. These 2019 results suggests that habitats downstream of the falls are used by the Arctic Grayling, while the more sensitive ddPCR results from 2021 suggest Arctic Grayling are absent upstream of the falls. Failure to find Arctic Grayling fry at a single electrofishing site near the stream mouth suggests that fry are rare or possibly absent in this cold stream (7.2°C, Table 2). Further sampling effort directed at adults might inform the eDNA detection of Arctic Grayling downstream of the falls.

Remarkably, fish sampling in Trib-4 found an important bull trout population sustained within 1.5 km of stream habitat situated between two impassable falls (Figure 11). Headwater exchange and isolation of populations upstream of impassable falls provide opportunities for further studies of ecology (e.g., critical habitat area that sustain

populations). The reproductive isolation from downstream also promotes rapid evolutionary divergence and unique local adaptations (Taylor 1999; McPhail 2007). Consequently, the isolated above barrier Bull Trout population discovered in Trib-4 is a unique and important component of diversity among Ospika Bull Trout and should be highlighted for conservation management.

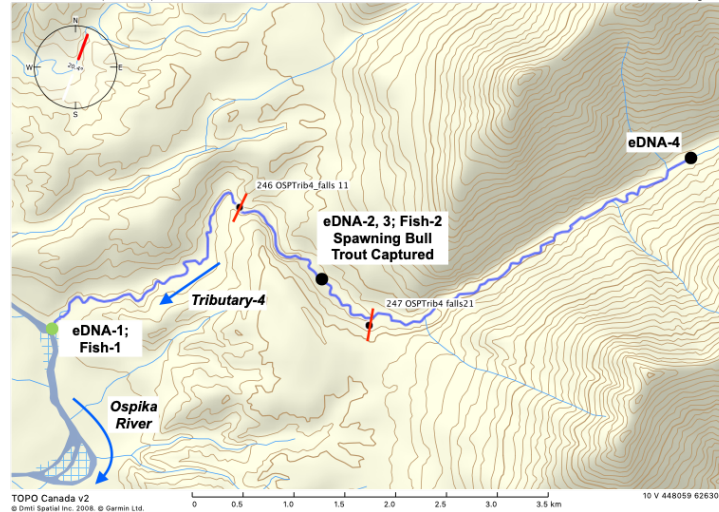


Figure 11: Sampling in unnamed Tributary 4 between 2019 and 2021. Labelled dots indicate fish and eDNA sample site locations (Green = Arctic Grayling eDNA detected; Black= Arctic Grayling eDNA not detected). Blue arrows indicate direction of stream flow, red slash indicate impassable (5meter high) falls.

4.5.3 Aley Creek (eDNA site Trib-3).

Three fish sampling sites sampled 2.2 km of fish habitat (Table 4) distributed within 12 km of Aley Creek and failed to capture Arctic Grayling. All fish were captured at two electrofishing sites that sampled just over 364 m in downstream reaches and no fish were angled upstream. The detection of Arctic Grayling eDNA at the mouth in 2019 together with results from fish sampling indicate that Arctic Grayling share habitat with Bull Trout and Slimy Sculpins (Table 4) in the extensive riffle pool habitats. Cold temperature and absence in electrofishing sites suggests, however, that the eDNA detected adult Arctic Grayling that requires confirmation with further sampling (angling) effort.

4.5.4 Finlay Reach streams

Electrofishing and angling efforts were spread thinly among seven sites in the four Finlay Reach streams and, unfortunately, absence of Arctic Grayling provide limited information about the habitat use upstream of eDNA detections. Electrofishing sites in Chowika Creek and Davis River were like those sampled by Langston and Blackman (1993), and sampled extensive low gradient areas situated, respectively, 12 and 8 km upstream from the mouth. Also, like Langston and Blackman (1993), sampling failed to inform the hypothesis that natal areas might exist in these fifth order streams. Absence of Arctic Grayling in the fish sampling supports the hypothesis that Arctic Grayling are not present in the vast low gradient areas situated in the upstream reaches of Chowika Creek.

Historically in 1989, Langston and Blackman (1993) used open site electrofishing to sample similar low gradient sites in the same Finlay Reach streams and captured similar numbers and almost the same fish species (Table 7). Sampling in 2021 covered more than 10 times the stream distance, possibly in different river conditions, and using methods biased toward capturing Arctic Grayling. Consequently, fish densities are not comparable between 2021 and 1989. Absence of Arctic Grayling both in 1989 and 2021 suggests, however, that the eDNA detected low densities of Arctic Grayling and the results do not support the hypothesis that Arctic Grayling abundance is increasing. Furthermore, more intensive sampling is needed to identify the life histories present and address the hypothesis that these streams provide more than just adult rearing areas (i.e., natal areas or yearling habitats). Fish sampling in 2021 evaluated 2.9 km of stream habitat in Finlay Reach streams, encompassing only 40% of the stream lengths evaluated by eDNA and with lower capture efficiency (Table 4).

Table 7: Comparisons of August fish capture from similar reaches in electrofishing surveys historically (one site per stream; Langston and Blackman 1993) and most recently (in this 2021 survey) among the same Finlay Reach watersheds. Longer sample lengths in 2021 reflect multiple sites in Collins and Lafferty creeks and using sampling methods biased toward capturing Arctic Grayling. CCG= Slimy Sculpin; RB= Rainbow Trout; BT= Bull Trout; MW= Mountain Whitefish; LSU= Longnose Sucker.

Finlay Reach Stream	Electrofishing in 1989			Electrofishing in 2021		
	Length Sampled (m)	#Fish Captured	Species	Length Sampled (m)	#Fish Captured	Species
Lafferty Creek	50	37	CCG, BT, RB	622	32	CCG, RB, BT
Collins Creek	50	12	CCG, BT, MW	650	30	CCG, BT
Davis River	50	34	CCG, BT, LSU	450	4	BT, MW
Chowika Creek	75	26	CCG, BT	300	30	BT

Failure to capture Arctic Grayling in angling and electrofishing efforts in Collins and Lafferty creeks suggests that the five eDNA sites in these streams detected low densities of Arctic Grayling. Unfortunately, fish sampling failed to inform the hypothesis that the extensive sinuous low gradient habitats provide habitats for adults and yearlings, and potentially fry.

5.0 Conclusions and recommendations

5.1 Conclusions:

Ospika River and the four adjacent Finlay Reach streams are all cold, steep, and active streams and not typical among other Arctic Grayling core areas in upper Peace Basin. Typically, Arctic Grayling metapopulations thrive in watersheds that provide clear and complex stream habitats upstream for adult rearing, and low gradient larger habitats in downstream reaches provide rearing for fry and yearlings, and overwintering habitats (e.g., Parsnip Core Area, Blackman and Hunter 2001). Young-of-the-year emerge from spawning sites and fry seek out local quiet warm areas to rear along stream margins of large rivers and tributary confluences (reviewed in Stamford et al. 2017). As they grow Arctic Grayling fry shift their habitats to include increasingly swifter flowing waters and in some Peace Basin core areas, appear to move into large river habitats by around

60mm size (e.g., Nation River, Cowie and Blackman 2007). The sizes of fry captured using beach seine in other large rivers (e.g., Parsnip River, Upper Finlay River) have revealed, however, that the main channels are also important rearing habitats for smaller sized fry (Blackman and Hunter 2001; Figure 12).

Seining almost three kilometers of habitat in Ospika main channel has provided the first confirmed Arctic Grayling fry record in the watershed. This first evidence for the presence of natal areas supports the hypothesis that Ospika River and adjacent Finlay Reach streams support an independent metapopulation. Further investigations are needed, however, to evaluate the critical habitats, limiting factors, and threats to this apparently rare population.

The capture of only two large (60mm) fry suggests that Arctic Grayling natal areas might be less connected to the main channel than in Parsnip and Finlay rivers. The captured fry appear to have dispersed from quiet areas and might signify the importance of the off-channel, and lower reaches of tributaries as core habitat areas for Arctic Grayling. The cold temperatures, swift flows, abundance of predatory bull trout, and potentially interactions with Kokanee, appear to be limiting factors that constrain habitat use into small patches in Ospika River. Possibly, fry abundance is higher where warm groundwater and tributaries provide both rapid growth rates, and a refuge from cold adapted predators. Possibly, turbid conditions associated with Ospika River provide cover for rearing fry and an escape from predation. All life history stages of Arctic Grayling populations sustained in turbid streams have received limited study and are probably more frequent than is currently thought (e.g., Butcher et al. 1981; Stewart et al. 1982).

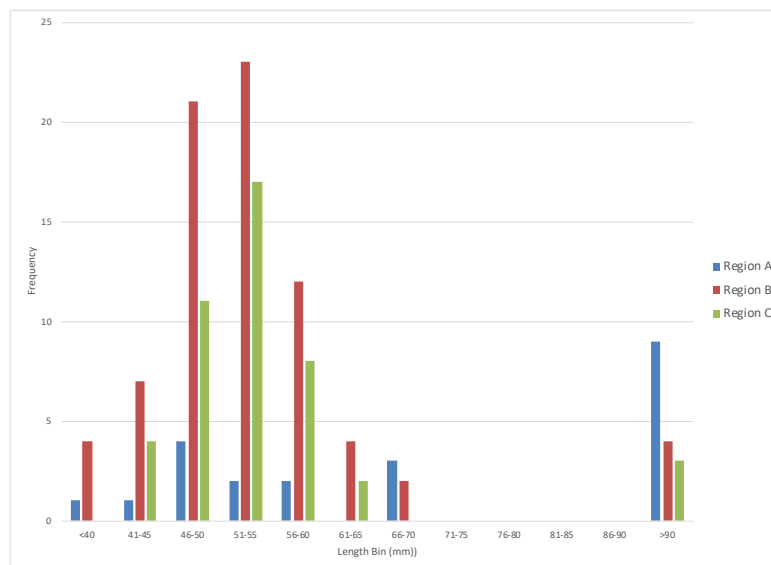


Figure 12: Length frequency distribution of Arctic Grayling fry captured during August 2017 in beach seine hauls in Finlay River (Upper Finlay Core Area). Fry were distributed along 50 km of main channel habitat. Coloured bars indicate three regions listed from upstream (Region A) to downstream (Region C). Data from Chu Cho 2017.

Although interactions between Arctic Grayling and native fluvial Kokanee have long influenced the adaption and habitat use between the species in Finlay Reach streams

(e.g., Fielden 1991, 1992), the recent introduction of adfluvial Kokanee and their recent abundance increase in Ospika River is a possible threat (Wilson and Shrimpton 2020). Increased competition among fry in combination with a concordant increase in predation from Bull Trout might be seriously impacting this apparently rare Arctic Grayling populations.

There are at least four reasons why it is important to continue investigating the Arctic Grayling in Ospika River.

1. Persistent detections of Arctic Grayling eDNA in Finlay Reach streams suggest Ospika River and adjacent Finlay Reach streams are unique among other streams in Williston and Upper Peace core areas where Arctic Grayling eDNA is absent.
2. Presence of fry in the fish sampling supports the hypothesis that Ospika River sustains an independent metapopulation, but further sampling is required to define the critical habitats.
3. Rare and potentially uniquely adapted populations provide significant function to ecosystems and improve the chance of the species survival in the face of environmental changes.
4. Finlay Reach Arctic Grayling appear to have survived flooding and potentially contain those life history characteristics best suited to cope with the changing environmental conditions and interactions with fish assemblages (e.g., Bull Trout Predation) in Williston Reservoir. Hypothetically, a unique independent metapopulation with natal areas in Ospika River complete their life history by migrating among adjacent streams through the reservoir to rear in watersheds dominated by Bull Trout.

5.2 Recommendations:

1. Continue sampling to determine the distributions of critical natal areas in Ospika River, focusing sampling efforts in quiet off-channel habitats and tributaries influenced by warm inflows. Draw from sampling results (i.e., Figure 9) to focus locations of sampling efforts.
2. Increase sampling effort for adults and yearlings to identify critical habitats in all watersheds and help inform migratory behaviour, including knowledge from the local sports fishery.
3. Added eDNA sampling will improve distribution data in Finlay Reach streams. For instance, address the hypothesis that Arctic Grayling distribution is constrained to the downstream reaches by abundant Bull Trout, similar to Davis River.
4. Collect genetic (tissue) and age structure samples from all life history stages to enable evaluations of migratory behaviour and possible connections with surrounding core areas by comparing with previous genetic studies (i.e. Stamford and Taylor 2005; Shrimpton et al. 2012).
5. Evaluate threats to this apparently small population from changing fish assemblages (e.g., introduced kokanee abundance and associated increased predation from bull trout) and resource developments (e.g., mining, forestry).

6.0 Literature

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