

Metal Concentrations in an Arctic Wastewater Wetland: Insights and Innovations from Baker Lake, Nunavut, Canada

by
Bronte E. McPhedran

A thesis
presented to the University of Waterloo
in fulfillment of the
thesis requirement for the degree
of Master of Science
in
Biology

Waterloo, Ontario, Canada, 2024

©Bronte E. McPhedran 2024

Author's Declaration

I hereby declare that I am the sole author of this thesis. This is a true copy of the thesis, including required final revisions, as accepted by my examiners. I understand that my thesis may be made electronically available to the public.

Abstract

The unique climatic conditions and isolated geographies of Arctic communities present distinct challenges for municipal wastewater treatment. Most communities depend on natural wetlands to provide passive treatment of wastewater; however, increased pressure on these systems from prolonged use, climate change, and intensifying human activity has resulted in the recent development of northern-specific standards for wastewater. These standards, among other objectives, aim to maintain healthy fish populations and diverse aquatic ecosystems, while calling for community consultation, integration of traditional knowledge, and adherence to federal and territorial regulations. A passive wastewater treatment system near Baker Lake, NU, served as a case study to address knowledge gaps regarding ecosystem function and trace metal dynamics in abiotic and biotic ecosystem compartments subject to wastewater inputs. Additionally, this research explored a method for time-resolved analysis of contaminant histories, which is particularly valuable in environments lacking baseline data or where regular monitoring is challenging, including in the Arctic.

Between the summers of 2019 and 2021, a series of lakes that represented wastewater-affected and reference systems were sampled for water, sediment, and fish. Water from wastewater-affected lakes had elevated concentrations of metals (e.g., Mn, Fe, Cu) and increased primary productivity (as inferred from chlorophyll-*a* concentrations), with persistent hypereutrophic conditions; extremely low dissolved oxygen levels were recorded in the lake closest to wastewater influent. Although sediment analysis near wastewater sources indicated higher levels of Cu and Zn, overall sediment chemistry was similar across all sites. The influence of wastewater on the trophic ecology of Burbot (*Lota lota*) was minimal, whereas Arctic grayling

(*Thymallus arcticus*) had more enriched (i.e., less negative) $\delta^{13}\text{C}$ at wastewater-affected sites, suggesting altered energy pathways in lakes that receive nutrients from wastewater. Analyses of metals in muscle tissue revealed lower, although not significant, concentrations of methylmercury – the toxic, biomagnifying form of mercury - in fish from wastewater-affected sites compared to those from reference sites. These lower concentrations of methylmercury in fish muscle tissue occurred despite higher concentrations in abiotic media, potentially because of somatic growth dilution, but more research is necessary. Analyses of trace metals in otoliths revealed distinct multi-element otolith signatures (i.e., Zn, Mn, Cu, Fe, and Pb) between wastewater-affected and reference lakes, and significantly greater concentrations of Fe in a lake receiving wastewater across two reconstructed periods of growth. Together, findings from this research not only fill knowledge gaps regarding the influence of municipal wastewater on cold, oligotrophic freshwater systems in the subarctic, but also highlight the potential uses and limitations of otoliths as markers of wastewater exposures.

Acknowledgments

I extend my deepest gratitude to Dr. Heidi Swanson, my co-supervisor, mentor, and friend. Your enthusiasm for research, combined with your unwavering commitment to doing what is right, regardless of external pressures, is truly inspiring. I am profoundly grateful for your mentorship and guidance over the years. Thank you for taking me under your wing, even when I was unproven in my capabilities. You provided a space for me to grow as a field ecologist, scientist, and individual, and for that, I am forever grateful.

Thank you to my co-supervisor, Dr. Mark Hanson, for encouraging my participation in scientific conferences and for your memorable support during our fieldwork in Baker Lake—especially for keeping the Peregrine Falcon at bay while I set minnow traps. I appreciate your gentle encouragement and steadfast support.

Dr. Mark Servos, I am honored to have you on my committee. Your adaptability and scientific excellence during the challenges of the COVID-19 pandemic have been truly inspiring.

Thank you to Dr. Panseok Yang and Dr. Norm Halden from the University of Manitoba for your expertise in LA-ICP-MS and for the use of your facilities. Additionally, I extend my gratitude to all members of the Biotron (University of Western Ontario) and BASL (University of Alberta) laboratories for their contributions to sample analyses.

My endless gratitude goes to the community of Baker Lake. Thank you for the opportunity to immerse myself in your diverse aquatic life, flora, and fauna, and for the privilege of studying your beautiful land, which led to this thesis. Ma'na Kathy and Mary Susan for your cheerful greetings each morning. I am also thankful to Karen Yip from the Hamlet Council, along with Jamie and Valarie of the KIA, for your engaging conversations and support on our projects.

I thank Daniel Tapatai for your remarkable skills; I eagerly anticipate seeing you achieve your dreams in videography and photography. A big thank you to the participants of our 2023 sampling workshops, it was wonderful getting to know you all: Linda, Tara, Bosh, Mike, Damion, and Jamiya. And a special shout out to Derek Nateela for his assistance over the years. From the many trips on the all-whether road, with field crew or samples, to helicopter rides and picking berries—thank you for all of it.

Special thanks to the Agnico Eagle Environment Department for ensuring the success of my field work, providing daily morning stretches, and safety check-ins. I am particularly thankful to Leilan Baxter and Marie-Pier Marcil for your logistical support, and to Ryan VanEngen for your strategic guidance and encouragement, which was crucial for our 2021 fieldwork.

Thank you to the current and former members of the Swanson Lab for your support and enriching discussions. Special mentions to Amy Nguyen for your administrative and laboratory assistance, and Kirsten Nikel for being an amazing teammate during our lab transitions.

I have had incredible help with field work. First, I extend my thanks to Jared Ellenor. I am filled with gratitude for our fieldwork together; your early lessons in perseverance and secondary fun are fundamental. And now, recorded for posterity, you won the pasta bet. Thank you, Noel, for your field expertise and overcoming challenges. Madeline Prater, Jiminy Cricket, you are simply the best; thank you for your abundance of positivity and wit.

Finally, I would like to thank my family and my cat, Marcus. To my friends and loved ones, I am grateful for your support, encouragement, laughter, and love as I ventured into the realm of academia. Thank you for watching me evolve into the person I am today.

This research was made possible through funding and in-kind support from Agnico Eagle Mines Limited, an NSERC Collaborative Research and Development Grant (to R. Jamieson, Dalhousie University), NSERC Discovery and Northern Research Supplements, and a Province of Ontario Early Researcher Award (Swanson). Additional support was provided by Mitacs Training Program (2020) during restrictions from the global pandemic, the Northern Scientific Training Program for fieldwork (2021), and the NSERC Student Ambassador grant for community engagement (2022). Thank you also to the Tumkur family for their generous scholarship (2022) in honour of their children, Rammohan and Chitrakleha.

Dedication

To queer youth, past, present, and future. This work is dedicated to the strength and resilience that defines each journey, which has inspired and guided my own path in research.

Table of Contents

Author's Declaration.....	ii
Abstract	iii
Acknowledgments.....	v
Dedication.....	vii
List of Tables.....	x
List of Figures.....	xii
List of Symbols and Abbreviations	xiv
Chapter 1 General Introduction	1
<i>1.1 Treatment of municipal wastewater in the Canadian Arctic.....</i>	<i>1</i>
<i>1.2 Downstream effects of wastewater on water quality and biota.....</i>	<i>4</i>
<i>1.3 Monitoring Arctic freshwater ecosystems</i>	<i>6</i>
<i>1.4 Local context</i>	<i>9</i>
<i>1.5 Study rationale.....</i>	<i>11</i>
<i>1.6 Study Objectives.....</i>	<i>12</i>
Chapter 2 Metal Dynamics in a Natural Lagoon-Tundra Wastewater Wetland: Insights from Baker Lake, Nunavut, Canada.....	14
<i>2.1 Introduction.....</i>	<i>14</i>
<i>2.2 Methods</i>	<i>19</i>
<i>2.3 Results and Discussion</i>	<i>29</i>
<i>2.4 Conclusion</i>	<i>55</i>
Chapter 3 Beyond Soft Tissues: Otolith Microchemistry as a Potential Monitoring Tool in Arctic Freshwater Ecosystems	58
<i>3.1 Introduction.....</i>	<i>58</i>
<i>3.2 Methods</i>	<i>63</i>
<i>3.3 Results and Discussion</i>	<i>71</i>
<i>3.3 Conclusion</i>	<i>81</i>

Chapter 4 General Discussion	83
4.1 Research Synthesis	83
4.2 Limitations and Study Design.....	85
4.3 Future work	87
4.4 Relevance to the Hamlet of Baker Lake and the wider Canadian Arctic	88
4.5 Concluding Remarks.....	89
References	90
Appendices	107
Supplementary information for Chapter 1.....	107
Supplementary information for Chapter 2.....	108
Supplementary information for Chapter 3.....	117
Supplementary information for Chapter 4.....	126

List of Tables

Table 2.1 Comparison of water chemistry parameters between wastewater-affected and reference sites using Welch's t-tests, with false discovery rate adjustments for multiple comparisons. In wastewater-affected sites, concentrations of Mn, Fe, Cu, and Pb were significantly greater than reference sites. Concentrations of Zn, Ba, and MeHg were not significantly different between site-types, although MeHg was limited to one year of data due to sample contamination. Lake productivity parameters (i.e., chl _a , TN, and TP), as well as conductivity were significantly higher in wastewater-affected sites. There was no significant difference in dissolved oxygen (DO) concentrations between site-types, though low DO concentrations were noted in Lagoon Lake. When compared against the Canadian Council of Ministers of the Environment (CCME) guidelines and Federal Water Quality Guidelines (FEQG) for the protection of freshwater aquatic organisms, no exceedances were observed for dissolved metal concentrations. However, DO levels in Lagoon Lake were below acceptable CCME guidelines for all life stages.	32
Table 2.2 The 2019 and 2020 average Trophic State Indices (TSI) calculated for each lake from three index variables: chlorophyll- <i>a</i> (chl _a), total phosphorous (TP), and Secchi depth (SD). Equations are as follows: $TSI_{(chl_a)} = 9.81 \ln(chl_a) + 30.6$, $TSI_{(TP)} = 14.42 \ln(TP) + 4.15$, and $TSI_{(SD)} = 60 - 14.41 \ln(SD)$. TSI Classification: oligotrophic (O): mesotrophic (M), eutrophic (E), hypereutrophic (HE). Values are means ($\pm$ SE).	34
Table 2.3 Comparison of sediment chemistry between wastewater-affected and reference sites using Welch's t-tests with corrections for false detection rates. Significant differences in copper and zinc concentrations were found. Mean sediment concentrations were compared to guidelines set by the Canadian Council of Ministers of the Environment (CCME) when available. Copper concentrations in Lagoon Lake were between the threshold effect level (TEL) and probable effect (PEL) level, signifying that negative effects on biota occasionally occur at similar concentrations.	42
Table 2.4 Summary of results from Linear Models (LM) and Linear Mixed Models (LMM) assessing differences between wastewater-affected and reference sites in trophic ecology (i.e., $\delta^{15}N$, $\delta^{13}C$), energy storage (i.e., C:N, condition), and growth of Arctic grayling. The random effect of lake was tested for each covariate. If variance estimates approached zero, and singularity was detected, the model was deemed to be overparameterized. In such instances, relationships were analyzed using a linear model (LM) without a random effect. Parameter estimates (β), standard errors (SE), degrees freedom (df), the test statistic (i.e., χ^2 ; chi-square or F; F value), and significance (p) are shown. Significant differences ($\alpha = 0.05$) are underlined and bold.	47
Table 2.5 Summary of results from Linear Models (LM) and Linear Mixed Models (LMM) assessing differences between wastewater-affected and reference sites in trophic ecology (i.e., $\delta^{15}N$, $\delta^{13}C$), energy storage (i.e., C:N, condition), and growth of Burbot. The random effect of lake was tested for each covariate. If variance estimates approached zero, and singularity was detected, the model was deemed to be overparameterized. In such instances, relationships were analyzed using a linear model (LM) without a random effect. Parameter estimates (β), standard errors (SE), degrees freedom (df), the F-value (F), and significance (p) are shown. Significant differences ($\alpha = 0.05$) are underlined and bold.	48

Table 2.6 Results of Linear Models (LM) and Linear Mixed Models (LMM) comparing metal concentrations in muscle tissue from A) Arctic grayling and B) Burbot between wastewater-affected and reference site-types from Baker Lake, Canada, Nunavut. The adjusted R^2 ($R^2_{(adj)}$) describes the proportion of variance explained once the number of predictors is accounted for. The marginal R^2 ($R^2_{(m)}$) describes the proportion of variance explained by fixed factors and the conditional R^2 ($R^2_{(c)}$) describes the proportion of variance explained by both fixed and random factors. For LM, the test statistic is the F-value (F), and for LMM, it is the chi-squared (χ^2) value. Significance level (α) is set at 0.05.	52
Table 2.7 Results of a multiple linear regression (MLR) analysis examining the influence of fish life history and trophic ecology variables on methylmercury concentrations ([MeHg]) in a) Arctic grayling and b) Burbot muscle tissue from wastewater-affected (n=3) and reference (n=2) sites. Variation in [MeHg] attributed to fork length was accounted for by using residuals from a preliminary regression. Covariates were included based on criteria set by tolerance (Tol.) and variance inflation factors (VIF) to minimize multicollinearity. The table reports the F-values and significance levels for each predictor.....	55
Table 3.1 Mean $\pm$ standard error metal concentrations (in parts per million; ppm) in otoliths of Arctic grayling from three lakes near Baker Lake, NU. Metal concentrations are reported for two regions of otolith growth: 1) year-one (i.e., first full year of growth after hatch) and 2) marginal edge (i.e., last full year of growth before capture). Marginal edge values are corrected for body size by adding residuals of the length – [metal] linear regression to mean [metal]. Significant pairwise differences of ($\log_{10}$ -transformed) metal concentrations are depicted with compact letter display (Tukey's HSD on ANOVA, $p < 0.05$) and bold. ANOVA results reported in Table S3.8.	74

List of Figures

- Figure 2.1 Map of the study sites in Baker Lake, Nunavut, Canada. 20
- Figure 2.2 Principal component analysis (PCA) biplot of water quality parameters in wastewater- affected (triangle) and reference (square) lakes. Individual lakes are labeled on the plot. Results for Upper Lagoon Lake (UL; Circle) are included for information only and were not used to determine eigenvalues. Dimension 1 (Dim1) of the PCA explains 62.4% of the variation among lakes and is highly correlated with several analysed metals. Dimension 2 (Dim2) explains an additional 19.6% of variation among lakes and is highly correlated with parameters that indicate primary productivity, including Chla, DOC, and SO_4^{2-} , which are responsible for 24.3%, 19.6%, and 10.0% of Dim2 variation, respectively. Lakes closest to wastewater input (i.e., Lagoon and Finger Lake) were separated from reference lakes across Dim1, indicating high contributions of metal concentrations contribute to dissimilarity. The furthest downstream wastewater lake (i.e., Airplane Lake) separates from reference lakes on Dim2 more than Dim1, indicating that metal concentrations are more similar to reference sites in this furthest downstream lake. 31
- Figure 2.3 Arithmetic mean ($\pm$ SE) values for water quality parameters, using all available data (2019 and 2021 and all technical replicates). Note that for MeHg only data from 2021 were available. Wastewater-affected sites (i.e., Lagoon Lake, Finger Lake, and Airplane Lake) had higher mean values compared to reference sites (i.e., R1 and R2) for all parameters except DO. DO in Lagoon Lake was below the lowest acceptable concentration for protection of aquatic life in Lagoon Lake (CCME). Upper Lagoon Lake (UL) is directly upstream of the wastewater wetland and is included for visual comparison only. 35
- Figure 2.4 Photographs from 2019 field work in Baker Lake, NU showing: a) a settlement pond adjacent to the wastewater wetland, with a visibly damaged gravel berm that allows effluent to bypass directly into the wetland, and b) the inflow of effluent from Lagoon Lake to Finger Lake, facing west. The water is notably turbid and large amounts of periphyton are visible on submerged boulders. 36
- Figure 2.5 Principal component analysis (PCA) biplot of sediment parameters in wastewater-affected (triangle) and reference (square) sites. Individual lakes are labeled on the plot, including Upper Lagoon Lake (UL; Circle) which is provided for information only and was not used to estimate eigenvalues. Dimension 1 (Dim1) of the PCA explains 46.8% of the variation among lakes and is highly correlated with concentrations of Mn, THg, and Fe. Dimension 2 (Dim2) explains an additional 32.3% of variation among lakes and is highly correlated with concentrations of Zn, Cu, and MeHg. Lakes closest to the wastewater input (i.e., Lagoon and Finger Lake) differ from reference lakes across both dimensions, as influenced by [MeHg] and [Cu], respectively. The final wastewater Lake (i.e., Airplane Lake) and R1 have similar positions on Dim1, therefore, variation may be explained by parameters strongly correlated with Dim2 (Zn, Cu, MeHg). Airplane Lake differs from R2 across both dimensions. 38
- Figure 2.6 Arithmetic mean ($\pm$ SE) values for sediment metal concentrations, using all available data (2019 and 2021 and all technical replicates). A distinctly high concentration of MeHg was measured in Lagoon Lake compared to other impacted lakes and non-impacted lakes. THg measured in all study lakes was well below the CCME freshwater guideline (PEL: 0.486 $\mu\text{g/g}$ dry weight) for the protection of aquatic life..... 40

- Figure 2.7 Isotope biplot of length-adjusted mean $\pm$ standard error $\delta^{15}\text{N}_{\text{adj}}$ and $\delta^{13}\text{C}$ for Arctic grayling and Burbot from wastewater-affected sites ($n=3$; green triangles) and reference sites ($n=2$; blue squares) near Baker Lake, Nunavut, Canada. A significant difference in Arctic grayling $\delta^{13}\text{C}$ was observed ($LM, F(1, 60) = 9.475, p = 0.003$), but not in $\delta^{15}\text{N}_{\text{adj}}$ or either isotope for Burbot. $\delta^{15}\text{N}_{\text{adj}}$ values are mathematically corrected for baseline values in each lake. 44
- Figure 2.8 Box and whisker plots of size-corrected metal concentrations ($\mu\text{g/g ww}$; wet weight) $\pm$ standard error in muscle tissue for Arctic grayling (ARGR) and Burbot (BURB) from reference (REF, blue squares) and wastewater-affected (WW, green triangles) systems. Linear models (LM) and Linear Mixed Models (LMM) were used to test site-type differences. To account for the effect of body size, residuals from the length – metal concentration relationship were calculated and adjusted to the mean length before modeling. Lake was included as a random factor (wastewater-affected: $n=3$ lakes; reference: $n=2$ lakes); when singular bounds were detected, the random factor was removed to avoid over-parameterization. Muscle tissue metal concentrations were significantly lower in Arctic grayling ($LMM, \chi^2(1) = 11.102, p < 0.001$) and Burbot ($LMM, \chi^2(1) = 4.299, p = 0.038$) from the wastewater-affected systems compared to reference systems. Significant difference are signified by an asterisk (*) with alpha set at 0.05. 53
- Figure 3.1 Study lakes in Baker Lake, Nunavut, Canada. A subset of three lakes were sampled for otolith microchemistry analysis, including Upper Lagoon Lake, Airplane Lake, and Reference 1 (R1). 64
- Figure 3.2 Principal component analysis (PCA) biplots for a) year-one otolith signatures and b) marginal edge otolith signatures. Each point on the plot signifies an individual fish metal signature based on measurements of five metals (Fe, Pb, Mn, Cu, and Zn) quantified using ICP-MS. Study lakes include Upper Lagoon Lake (circles), upstream of wastewater input; Airplane Lake (triangles), receives wastewater; and, the reference, Lake R1 (squares). Elemental signatures of both year-one and marginal edge differed significantly among the fish from the three study lakes, although marginal edge otolith composition ($PerMANOVA, F(2, 50) = 4.1822, p = 0.002$) was more distinct than year-one ($PerMANOVA, F(2, 48) = 4.038, p = 0.012$). Lead (Pb) contributed the most to among-lake variance in metal signatures for both year-one and marginal edge otolith regions. 72
- Figure 3.3 Plots of Arctic grayling otolith Zn and Mn concentrations (ppm) smoothed ($n=10$) and plotted over distance (μm). Oscillatory patterns of [Zn] and [Mn] align with annular growth structure in otoliths from both: a) a lake downstream of wastewater input (i.e., Airplane Lake) and b) a reference lake not connected to the wastewater wetland (i.e., Lake R1). 78

List of Symbols and Abbreviations

*	Multiply
‰, ppm	Part per million
°C	Degrees Celsius
Airplane	Airplane Lake
Al	Aluminum
As	Arsenic
BL Creek	Baker Lake Creek
C:N	Elemental bulk tissue carbon to nitrogen ratio
Ca	Calcium
CBOD ₅	Carbonaceous biochemical oxygen demand
Cd	Cadmium
Cl-	Chloride
Cu	Copper
DO	Dissolved Oxygen
DOC	Dissolved organic carbon
EEM	Environmental Effects Monitoring
Fe	Iron
Finger	Finger Lake
Hg	Mercury
Lagoon	Lagoon Lake
Log ₁₀	Logarithm base ten
mBar	Millibar (metric unit of pressure)
MeHg	Methylmercury
mg	Milligram
mm	Millimetre
Mn	Manganese
N ₂	Nitrogen gas
NH ₄	Ammonium
Ni	Nickel
<i>P</i>	Significance value
Pb	Lead
QA/QC	Quality assurance/Quality control
R1	Reference lake 1
R2	Reference lake 2
Se	Selenium
SiO ₂	Silica
SO ₄	Sulfate
Sr	Strontium
THg	Total mercury
TN	Total nitrogen
TP	Total phosphorous
TSS	Total suspended solids
WSER	Wastewater Systems Effluent Regulations
Zn	Zinc
δ ¹³ C	Delta fifteen carbon

$\delta^{15}\text{N}$	Delta fifteen nitrogen
$\delta^{15}\text{N}_{\text{adj}}$	Delta fifteen nitrogen adjusted for baseline values
μm	Micrometre
ICP-MS	Inductively coupled plasma mass spectrometry
LA-ICP-MS	Laser ablation inductively coupled plasma mass spectrometry
DMA	Direct mercury analyzer

Chapter 1

General Introduction

1.1 Treatment of municipal wastewater in the Canadian Arctic

People in communities of the Canadian Arctic have a unique connection to the land because they rely directly on it for wild-harvested foods and drinking water. In the 1950s, colonial policies in Canada resulted in Inuit - Indigenous inhabitants of what is now called the Canadian Arctic - changing from a semi-nomadic lifestyle to a lifestyle based in permanent human settlements. Human settlements imposed new requirements on local environments beyond subsistence, including receipt of year-round human waste. Waste management is a challenge for communities globally, and this is especially true in the Arctic, where extreme climates, remote locations, and minimal infrastructure limit the efficiency and efficacy of wastewater treatment ([Lam and Livingston, 2011](#)).

In response to persistent and widespread problems emerging from undertreated human wastewater in Canadian waterbodies, the Canadian government established the “Wastewater Systems Effluent Regulations” (WSER) under the Fisheries Act in 2012 (SOR/2012-139) ([ECCC, 2015](#)). The regulations were created to protect fish and fish habitat, and to prevent pollution. At the time of WESR implementation, the Northwest Territories, Nunavut, and communities north of the 54th parallel in Quebec or Newfoundland and Labrador were exempt from meeting the minimum effluent quality standards due to the unique, and often extreme, conditions and challenges in northern environments ([ECCC, 2015](#); [Johnson, 2008](#)). Municipal wastewater systems in exempt regions fall under the jurisdiction of provincial or territorial regulatory bodies.

For instance, wastewater projects in Nunavut are currently regulated by the *Nunavut Waters and Nunavut Surface Rights Tribunal Act* and are enforced by the *Nunavut Water Board*.

It is crucial to acknowledge the unique challenges of implementing efficient infrastructure in Northern communities, including the presence of permafrost, severe temperatures, costs of construction, and lack of trained personnel. This acknowledgment ensures that limited resources are not wasted on regulations that are tailored to more southerly regions and infeasible in the north. In Nunavut, the WSER exemption prompted discussions among territorial officials, Inuit organizations, community members, and researchers. They collectively agreed that understanding the current wastewater treatment systems in the Canadian Arctic is crucial before developing plans for future approaches ([Johnson, 2008](#); [Lam and Livingston, 2011](#)).

During the last decade, studies on wastewater treatment in the North have centered on design and function ([Schmidt et al., 2016](#); [Yates et al., 2020, 2012](#)), microbial risks ([Daley et al., 2022](#); [Gromala et al., 2021](#); [Hayward et al., 2018](#)), contaminants of concern ([Chaves-Barquero et al., 2016](#); [Stroski et al., 2020](#)), and public health implications ([Daley et al., 2018](#); [Health Canada, 2010](#)). With a deeper understanding of the operational constraints of wastewater treatment in the Arctic, a National Standard of Canada (CSA W203:19) was developed for the *Planning, design, operation, and maintenance of wastewater treatment in northern communities using lagoon and wetland systems*. This standard deems passive systems to be the most appropriate method for treatment of wastewater in Arctic environments.

Passive treatment in the Arctic dates back to 1969, following the introduction of guidelines for wastewater lagoons, also known as stabilization ponds ([Dawson and Grainge, 1969](#)). Lagoons are designed to hold community wastewater for an entire year. In winter,

wastewater accumulates, and heavy solids settle to the bottom. Treatment occurs from spring thaw to autumn freeze-up, typically spanning a period of three to four months ([Hayward et al., 2014](#)). During this ice-free period, engineered lagoons discharge water at scheduled intervals. In contrast, natural lagoons use existing tundra depressions, reinforced with gravel berms, to manage wastewater release ([Yates, 2012](#)). Natural lagoons are intended to gradually release water, but problems such as breaches in the containment berms or capacity overflows can lead to accidental releases. These incidents reduce the time wastewater stays in the lagoon, potentially undermining the effectiveness of the treatment process ([Balch et al., 2018](#)).

Wetlands naturally treat wastewater through a combination of physical, chemical, and biological processes. As wastewater flows through a tundra wetland, suspended solids settle, and microbes break down organic materials and nutrients ([Chouinard et al., 2014](#)). Additionally, plants take up nutrients, particularly nitrogen and phosphorus, and provide habitat for microbial communities that degrade contaminants ([Vymazal, 2007](#); [Yates et al., 2012](#)). Furthermore, water quality is improved through filtration, ultraviolet irradiation, chemical precipitation, and volatilization processes that occur within the tundra wetlands ([Balch et al., 2018](#)). A study conducted across four communities in Nunavut revealed that wetlands effectively reduced both carbonaceous biochemical oxygen demand (CBOD₅) and total suspended solids (TSS) by as much as 80% ([Ragush et al., 2015](#)). In a separate investigation in the same region, pharmaceutical concentrations were found to be at low levels, posing minimal risk to aquatic organisms ([Stroski et al., 2020](#)). However, the presence of per- and polyfluorinated alkyl substances raise concerns and signal the need for ongoing monitoring ([Muir et al., 2019](#)).

The effectiveness of passive wastewater treatment in wetlands depends on the relative rate of input and assimilation. For treatment to be successful, the rate of contamination should not surpass the rate of removal. While many Arctic wastewater wetlands have been operational for 30 to 40 years ([Yates, 2012](#)), research shows that newer and deeper systems are more effective, largely attributed to less resuspension of solids ([Ragush et al., 2015](#)). As such, Northern community members have raised concerns about the efficiency and capacity of existing wastewater systems. These concerns are heightened due to the introduction of new standards for Northern lagoon wastewater systems (CSA W203:19), increasing populations, and the emergence of new contaminants ([Johnson, 2008](#); [Yates et al., 2020](#)). Ultimately, residents in the North seek to understand and reduce the effects of municipal wastewater discharge on aquatic ecosystems and public health.

1.2 Downstream effects of wastewater on water quality and biota

Performance, and subsequent effluent quality from passive wastewater treatment systems in the Arctic, vary seasonally and are affected by loading and removal rates of primary constituents of interest, including nitrogen, phosphorus, and suspended solids ([Hayward et al., 2014](#); [Yates et al., 2012](#)). Wastewater contains a variety of contaminants including bacteria, viruses, heavy metals, and micropollutants such as pharmaceuticals, which often adhere to particulate matter. Previous studies, such as those conducted by [Balch et al. \(2018\)](#) and [Hayward et al. \(2014\)](#), have documented that the quality of effluent in tundra wetlands is at its lowest during the spring freshet when turbidity is often highest. Spring freshet often also results in decreased residence time, which reduces the efficiency of removing and transforming contaminants and excess nutrients from wastewater ([Stroski et al., 2020](#); [Yates et al., 2012](#)). In

short, physical properties of the wastewater and water flow vary seasonally, and thus so too does the efficacy of wetland treatment processes.

Other variables that impact performance of wastewater tundra wetlands include temperature of both water and soil, the diversity of plant and microbial communities, wetland surface area, and oxygen availability. [Huang et al., \(2021\)](#) found that biological processing and nutrient removal in Arctic wastewater lagoons was higher at warmer temperatures, likely due to elevated bacterial activity. Similarly, the treatment performance of lagoon-tundra systems reaches its peak at the end of open-water season, when plants exhibit the greatest capacity for nutrient assimilation and contaminant removal ([Yates et al., 2012](#)). In wetland soils where oxygen supply, or redox potential, is limited, microbial communities adapt by utilizing alternative electron acceptors, a shift that profoundly affects their composition and size. This adaptation is critical for processes like nutrient cycling and metal transformation, influencing the toxicity and bioavailability of various substances in the ecosystem.

Anoxia, a common occurrence in tundra wastewater wetlands, can begin during the ice-on season and persist even after ice break-up. A detailed study of a natural tundra wastewater wetland in Coral Harbour, Nunavut, reported anoxic conditions until mid-summer, which adversely affected treatment performance ([Hayward et al., 2014](#)). Low dissolved oxygen concentrations have significant impacts on local biota. In Iqaluit, Nunavut, for example, benthic invertebrate communities were found to be depauperate up to 580 m from the effluent source ([Krumhansl et al., 2015](#)). Similar negative effects of wastewater-induced anoxia on biota have been documented in southern Canada ([Chambers et al., 1997](#); [Tetreault et al., 2013](#)). Beyond the direct effect of anoxia, wastewater can further impair diversity, density, and composition of biota

through chronic exposure to contaminants, eutrophication, and habitat disturbances, such as changes in turbidity and temperature ([Chambers et al., 1997](#)).

1.3 Monitoring Arctic freshwater ecosystems

Arctic freshwater ecosystems, including rivers, ponds, wetlands, lakes, and streams, are integral to the nutrition, economy, and cultural practices of Inuit ([Hughson, 2010](#)). The subarctic Barrenlands, spanning mainland Nunavut and eastern portions of the Northwest Territories, are defined by dynamic networks of interconnected lakes and streams, known as chain-lake systems. These systems foster diverse ecosystems and influence regional hydrology, making them particularly sensitive to change in climate and land use ([Heino et al., 2020](#)). Comprehensive monitoring and management strategies are essential to effectively steward these ecosystems.

In Canada, various frameworks are used to monitor, assess, and protect ecosystem health. One such framework is the Canadian Environmental Effects Monitoring (EEM) program. Established as a robust and effects-based regulatory framework, the EEM has been instrumental in establishing monitoring criteria across multiple trophic levels ([Gibbons and Munkittrick, 1994](#); [McMaster et al., 2006](#); [Ribey et al., 2002](#)). Central to the EEM approach is the identification of significant impacts of environmental stressors through measurable biological effects in organisms. This framework, which effectively combines physical, chemical, and biological data, is applicable to a range of different ecosystems ([Munkittrick et al., 2010](#)). The Canadian EEM program exemplifies how targeted, multi-faceted approaches, such as choosing locally important species, can substantially contribute to our understanding and preservation of ecosystem health.

Generally, EEM programs focus on comparing fish and benthic invertebrates (i.e., representative of fish habitat) between receiving environments and references areas. The

selection of organisms for monitoring within the Environmental Effects Monitoring (EEM) framework is guided by the specific objectives and attributes of each study ([Kilgour et al., 2007](#)). Benthic invertebrates are particularly valued as indicators due to their limited mobility and relatively short life cycles. These characteristics enable them to provide rapid and localized reflections of environmental changes, offering timely insights into the health of aquatic ecosystems ([Kilgour et al., 2007](#)). Conversely, fish serve as comprehensive sentinels of ecological health, integrating and accumulating data from both abiotic elements, such as water quality and sediment conditions, and biotic factors, including the health of invertebrate populations ([Kilgour et al., 2005](#); [Munkittrick et al., 2010, 2002](#)). This integration makes fish exceptionally effective at signaling broader ecological shifts and disturbances.

A fundamental aspect of many environmental monitoring frameworks, including the EEM, is choice of sentinel species. A sentinel species is an organism that is particularly sensitive to environmental changes, making it a reliable indicator of the overall health of an ecosystem. Monitoring sentinel species, rather than relying solely on abiotic factors, such as water quality, help detect subtle yet significant ecological shifts, especially in response to pollutants or other anthropogenic pressures. [Munkittrick et al. \(2002\)](#) elucidate the advantages of choosing a species with a known home range or strong site fidelity. This approach minimizes variability within a site, leading to a monitoring protocol that is both robust and sensitive, with reduced natural bias.

Anthropogenic perturbations, such as those from industrial or municipal wastewater, impact biological endpoints that reflect energy use and storage, including age, growth, and liver or gonad size. Increased food availability from eutrophication can also affect population age structures, via increased rates of reproduction and survival rates, and faster growth rates

([Munkittrick, 1992](#)). Conversely, stressors that limit food availability may reduce energy use and storage. The complex interactions of multiple stressors, as well as the unique physiology and ecology of a sentinel species, result in varying effects for biological endpoints ([Fuzzen et al., 2016](#)). Within the EEM program, warning-level effects are identified by specific ecological thresholds, such as a greater than 25% difference in fish age, growth, and liver or gonad size between impacted and reference sites ([Munkittrick et al., 2009](#)). Additionally, [Kilgour et al. \(2007\)](#) state that a change exceeding 10% in fish condition factor, the ratio of body weight-to-length, is a significant indicator of ecological change. These criteria for warning-level effects in sentinel species within the EEM framework provide a basis for hypotheses development in this study of a wastewater wetland near Baker Lake, Nunavut, Canada.

1.3.1. Arctic grayling

Arctic grayling (*Thymallus arcticus*), or Sulukpaugaq in Inuktitut, is a widely distributed large-bodied fish species that is present in many circumpolar regions, including the Canadian subarctic and low Arctic. Highly valued by both subsistence and recreation fishers ([Kitikmeot Inuit Association, 2006](#); [Scott and Crossman, 1973](#)), Arctic grayling have plastic life histories ([Reed, 1964](#); [Scott and Crossman, 1973](#)) but are sensitive to environmental change, namely increased water temperature ([Stewart et al., 2007a](#)) and turbidity ([Birtwell, 1984](#)). As visual predators, turbidity influences feeding efficiency and diet, and contributes to Arctic grayling feeding opportunistically at intermediate trophic levels ([Stewart et al., 2007b](#)). Juveniles (< 150 mm FL) feed primarily on aquatic macroinvertebrates and are prey to larger grayling and top piscivores, such as Lake trout (*Salvelinus namaycush*) and Burbot (*Lota lota*) ([Stewart et al., 2007b](#)). The diet of adult Arctic grayling expands to include terrestrial insects and small-bodied fish as they

increase in size and age, yet adults remain prey for larger fish ([Stewart et al., 2007b](#)). The importance of Arctic grayling to subsistence and recreational fisheries, their sensitivity to anthropogenic change, and their generalist diet make them useful sentinel species (e.g., [Laske et al., 2018](#); [Reynolds et al., 1989](#)).

1.3.2. Burbot

Burbot (*Lota lota*), Tiktaalik in Inuktitut, sometimes referred to as ling cod, have a circumpolar distribution and occupy top trophic levels in many Arctic lakes ([Cott et al., 2011](#)). Primarily residing in benthic habitats, Burbot experience ontogenetic dietary shifts from macroinvertebrates to fish as they grow and mature ([Amundsen et al., 2003](#)). Once diet is primarily comprised of fish, prey size is dependent on body size ([Amundsen et al., 2003](#)). Despite some overlap with prey used by Arctic grayling early in life, Burbot occupy distinct trophic niches from Arctic grayling at most life stages. Thus, Burbot may be helpful as a secondary large-bodied sentinel species ([Munkittrick et al., 2002](#)).

1.4 Local context

The Hamlet of Baker Lake, or *Qamani'tuaq* (“where the river widens” in Inuktitut), is in the Kivalliq region of Nunavut, approximately 40 km northwest of the Kazan River, and 280 km inland from the western shore of Hudson Bay (*Tariuq*). In addition to the Kazan, the Thelon River is a major water source to Baker Lake, entering the lake immediately west of the Hamlet. The waterbody of Baker Lake drains through Chesterfield Inlet (*Igluligaarjuk*) into Hudson Bay.

Before the onset of European colonization, the Kazan River and surrounding drainage basin supported one of the few Inuit groups in the Canadian Arctic that lived inland ([Kigjugalik Webster, 2001](#)). Inuit peoples of the Barrenlands migrated with caribou herds, and supplemented

their diet with fish, berries, small mammals, and birds ([Kigjugalik Webster, 2001](#)). Complex circumstances in the early 20th century including famine, fur trade, and colonialist ideals resulted in the permanent settlement of nine different cultural groups on the northwest shore of Baker Lake ([Kigjugalik Webster, 2001](#))¹. Today, Baker Lake remains the only inland community in Nunavut. Over time, permanent settlement has imposed unprecedented challenges to Inuit who once lived semi-nomadic lifestyles ([Daley et al., 2015](#)). Waste management is one of these challenges.

In Baker Lake, sewage collected from private dwellings and municipal buildings is trucked to a dumpsite north of town, where it is deposited into an un-engineered (i.e., natural) holding cell that receives approximately 7.67×10^7 L of sewage per year ([Stroski et al., 2020](#)). The holding cell acts as a facultative pond in which solid waste settles to the bottom, and wastewater flows through a wetland floodplain down to Lagoon Lake, the first of three lakes in a passive wetland treatment system. A wetland connects Lagoon Lake to Finger Lake, which flows via a stream to Airplane Lake. With a slope of approximately 1-2%, the entire chain lake system is estimated to be 3.3 km in length, and 14-80 m in width. The community landfill is located on the south shore of Finger Lake and is a secondary source of contamination to Finger and Airplane lakes. In contrast to the generally clear waters observed on tundra landscape, Airplane Lake is turbid and visually green. Locals have altered their land use in response to the visual changes in Airplane Lake by ceasing all recreational and subsistence fishing activities for fear of contamination (P. Tapatai,

¹ According to Baker Lake elders the cultural groups of inland Inuit that settled on the northwest shore of Baker Lake include: *Iluiliqmiut*, *Kihlirniirmiut*, *Hanningayuqmiut*, *Ukkuhiksalingmiut*, *Qairniirmiut*, *Hauniqturmiut*, *Akilinirmiut*, *Harvaqtuurmiut*, and *Paarlirmiut* ([Kigjugalik Webster, 2001](#)).

pers. comm., Aug. 2019). Final discharge of effluent from the wastewater wetland occurs via a creek that connects Airplane Lake to Baker Lake.

The Lagoon-Finger-Airplane Lake wetland treatment area is situated 1 km north of the hamlet, within an 18 km² watershed, and has serviced municipal wastewater since at least 1980. In 2007, the Hamlet of Baker Lake hired private consultants to ensure the wastewater wetland effectively treated sewage and solid wastes. Consultants determined the holding cell was inadequate, with a retention time of only three days (Nunami Stantec, 2011). As a result, upgrades implemented in 2010 transformed the wetland into an "augmented natural wetland" (see Yates et al., 2020) by strengthening the holding cell walls, constructing an additional holding cell, and improving attenuation. Berms, comprised of coarse gravel and sand, were placed between the holding cells and Lagoon Lake as well as between Lagoon Lake and Finger Lake (Nunami Stantec, 2011). The Hamlet relies on the system improvements implemented in 2010 to protect downstream ecosystems and human health; however, berm failure is common during spring freshet². All publicly available water quality reports since 1994 have been summarized (Table S1.1) and indicate that exceedances of CCME guidelines are more common in June (freshet) than in July or August.

1.5 Study rationale

The lagoon-tundra wastewater treatment wetland adjacent to the hamlet of Baker Lake is part of a larger watershed that drains into the waterbody of Baker Lake, which is also the municipal water source for the Hamlet. Given that Baker Lake is a vital source of water for the

² Indicated by Water License Reports to the Nunavut Water Board (i.e., 2015, 2017), the Baker Lake Waste Disposal Foreman (P. Narkyagik, pers. comm., July. 2019), and personal field observations (i.e., 2018, 2019, 2021).

community, upgrades to the current wastewater treatment system have been requested by the community. Accordingly, Agnico Eagle Mines, which operates the Meadowbank Gold Mine located approximately 70 km north of town and employs many people in the community, has partnered with researchers, the Kivalliq Inuit Association, the Department of Fisheries and Oceans, and the Baker Lake Hunters and Trappers Organization to understand the current function and ecological state of the wetland treatment system surrounding Baker Lake.

Foundational research completed during the last decade has indicated that passive wastewater treatment is appropriate for Arctic regions; however, many of these studies did not address impacts on human and environmental health ([Daley et al., 2018](#)). Recent research has shown that systems engineered to regulate flow and allow a full season of passive treatment, combined with effective risk communication to communities, can mitigate risks to human health ([Daley et al., 2022, 2019](#)). While recent research has begun to assess and address the effect of municipal wastewater on Arctic marine ecosystems ([Back et al., 2021](#); [Schaefer et al., 2022](#)), the impact of wastewater on freshwater ecosystems has received limited research attention and remains a priority research area.

1.6 Study Objectives

The overall objective of this study is to improve the biological and ecotoxicological understanding of freshwater lagoon-tundra wastewater wetlands in the Canadian Arctic, using a case study of one system located near the Hamlet of Baker Lake, NU. The research conducted addresses community concerns regarding the efficacy of passive wastewater treatment via wetlands in the Baker Lake area and fills knowledge gaps regarding effects on fish. Responding to a common problem faced by researchers and communities alike in the Canadian Arctic with

respect to lack of pre-impact or longer-term baseline data, research was also conducted to assess whether otolith microchemistry can be used to reconstruct time-resolved concentrations of metals in fish from contaminated wastewaters.

To improve our understanding of freshwater lagoon-tundra wastewater wetlands in the Canadian Arctic, I: 1) characterized chemistry of abiotic and biotic ecosystem compartments along the wastewater flow path; 2) compared biological endpoints and metal concentrations of fish from wastewater-affected and reference systems; and 3) investigated metabolically inert fish otoliths as a tool for reconstructing past metal exposure. I hypothesized that 1) all ecosystem compartments from the wastewater-impacted lakes would have elevated trace metal concentrations relative to reference lakes; 2) nutrient concentrations and chlorophyll-*a* concentrations in water, and indicators of energy storage in fish (e.g., condition and lipid content), would be higher in wastewater-impacted lakes relative to reference lakes; and, 3) metal concentrations in areas of the otolith that represent recent growth would be higher in wastewater-impacted lakes relative to reference lakes, and would be correlated with concentrations of metals in soft fish tissue.

Chapter 2

Metal Dynamics in a Natural Lagoon-Tundra Wastewater Wetland: Insights from Baker Lake, Nunavut, Canada

2.1 Introduction

As the first inhabitants of the now Canadian Arctic, Inuit – meaning “the people” in Inuktitut – have always been deeply connected with their ancestral land ([Hughson, 2010](#)). Many Inuit rely on the land for food and water, making them acutely aware of, and vulnerable to, ecosystem degradation. The practice of harvesting traditional, or “country”, foods is not only vital for nutrition, but also central to many cultural and spiritual traditions ([Hughson, 2010](#)). Accordingly, this practice is a key aspect of land use autonomy for the Inuit ([Robin et al., 2021](#)). Country food in Northern Canada includes foods harvested by hunting and fishing (e.g., caribou, Arctic char), as well as foods gathered from the land (e.g., berries, eggs) ([Hughson, 2010](#); [Newell et al., 2020](#)). While the importance of country food highlights the deep connection Indigenous northerners share with their land, this relationship is increasingly challenged by changes in lifestyle and land use.

In the 1950s, government policies resulted in Inuit transitioning from semi-nomadic lifestyles to permanent settlements, which has resulted in more concentrated pressures on local environments, not only for subsistence foods and water, but also for wastewater management. In Arctic communities, wastewater is passively treated through natural or constructed wetlands via biogeochemical processes and dilution ([Yates et al., 2012](#)). Prolonged exposure to wastewater has caused noticeable ecological changes in these wetlands, such as greening and increased

vegetation. Residents of Arctic communities have expressed concerns about the potential long-term effects of wastewater on environmental and human health ([Daley et al., 2015](#)).

Observations of increased productivity in wastewater wetlands are indicative of eutrophic, or high nutrient, conditions caused by loading of nitrogen and phosphorus (e.g., [Wetzel, 2001](#)). Typically, Arctic lakes are mesotrophic or oligotrophic, with medium to low productivity. An influx of nutrients from wastewater can induce eutrophication and ultimately disrupt a number of ecosystem functions, with effects ranging from reduced water clarity and dissolved oxygen concentrations ([Holeton et al., 2011](#)) to impacts on health ([McMaster et al., 2005](#); [Tetreault et al., 2011](#)) and feeding behaviour ([Stewart et al., 2007b](#)) of fish. Additionally, eutrophication may enhance the bioavailability of contaminants, such as metals, further increasing risks to fish health and essential ecosystem services such as drinking water, recreation, and fisheries ([Skei et al., 2000](#); [Wetzel, 2001](#)).

In addition to causing eutrophication, the introduction of minimally treated municipal wastewater can lead to increased metal concentrations in aquatic environments. Bioavailability of metals loaded into receiving waterbodies is influenced by elemental speciation ([Tessier and Campbell, 1987](#)) and several water chemistry variables that are themselves affected by wastewater inputs, such as water hardness, pH, temperature, and concentrations of dissolved organic carbon ([Heath, 1995](#)). Metals in water exist as free ions, inorganic and organic complexes, and precipitates bound to organic matter ([Tessier and Campbell, 1987](#)). Metal uptake by aquatic organisms can occur via exposure from food and water ([Bryan, 1976](#)), and route of uptake affects tissue- and organ-specific concentrations and sequestration, which is an important consideration when monitoring contaminants in aquatic biota.

Sentinel fish species are often used to evaluate the impact of stressors on aquatic ecosystems because, as longer-lived organisms, fish generally reflect more extensive periods of exposure than lower trophic-level organisms (Parks et al., 1991). Sentinel fishes also integrate the effects of complex mixtures, such as municipal wastewater (Fuzzen et al., 2016) and are therefore relevant indicators of impacts on ecosystem health and functioning. By comparing biological endpoints, such as condition factor, of sentinel fish between contaminated and references areas, effects of stressors can be assessed (Kilgour et al., 2005; Munkittrick et al., 2002).

Quantifying trace metal concentrations in fish tissues is crucial for understanding how wastewater effluent affects aquatic ecosystems. This analysis helps assess the bioavailability of metals, which is key to understanding the risks they pose to higher consumers. Accumulation of diet-derived metals in fish tissue reflects the biogeochemistry of each metal and a myriad of other factors, including prey selection (i.e., bulk seston, fish) foraging habitat (i.e., benthic, pelagic), and trophic level (e.g., Campbell et al., 2005; Power et al., 2002). Foraging habitat and trophic level can be inferred from stable isotope ratios of carbon ($^{13}\text{C}/^{12}\text{C}$; $\delta^{13}\text{C}$) and nitrogen ($^{15}\text{N}/^{14}\text{N}$; $\delta^{15}\text{N}$), respectively, and relationships between metal concentrations and $\delta^{15}\text{N}$ in biota can also be used to quantify rates of biomagnification (Kidd et al., 1995b). A food web study in Baffin Bay demonstrated that biomagnification rates vary among trace metals (Campbell et al., 2005), with total mercury, methylmercury, and zinc biomagnifying between trophic levels as evidenced by their positive relationships with $\delta^{15}\text{N}$. In contrast, concentrations of cadmium, lead, and nickel were negatively related to $\delta^{15}\text{N}$ and therefore biodiluted. For some metals (e.g., copper, magnesium, selenium), concentrations were independent of trophic level (Campbell et al., 2005).

In addition to trophic position, metal concentrations in fish can vary between benthic and pelagic habitats (e.g., [Power et al., 2002](#)). In freshwater ecosystems, $\delta^{13}\text{C}$ are often used to indicate whether a fish predominantly forages on benthic or pelagic carbon sources ([Hecky and Hesslein, 1995](#)). [Hecky and Hesslein \(1995\)](#) found that top predators in oligotrophic lakes, such as those in the Arctic, have narrow $\delta^{13}\text{C}$ ranges due to dominant reliance on benthic-based carbon sources at all trophic levels. However, in eutrophic lakes with light-limited benthic zones, the range of $\delta^{13}\text{C}$ ratios in consumers may be larger due to reliance on both benthic (less depleted $\delta^{13}\text{C}$) and pelagic (more depleted $\delta^{13}\text{C}$) carbon sources ([Chandra et al., 2005](#)).

The influence of municipal wastewater on stable isotope ratios in organisms exposed to effluents is shaped by treatment level, final effluent quality, and receiving environment characteristics. Studies reveal that higher quality effluents are generally $\delta^{15}\text{N}$ -enriched ([Gaston and Suthers, 2004](#); [Hicks et al., 2017](#); [Robinson et al., 2016](#)), in contrast to raw or minimally treated wastewater, which is typically $\delta^{15}\text{N}$ -depleted ([Daskin et al., 2008](#); [deBruyn and Rasmussen, 2002](#); [Gaston and Suthers, 2004](#); [Hicks et al., 2017](#)). Enriched $\delta^{15}\text{N}$ results from longer nitrification periods during higher-level treatment. Other wastewater treatment processes, such as volatilization and denitrification, preferentially involve the lighter stable isotope of nitrogen and can also result in $\delta^{15}\text{N}$ enrichment ([Heaton, 1986](#)). $\delta^{15}\text{N}$ has been effectively used to trace assimilation of nutrients from municipal wastewater sources along a temperate river system; enriched values were consistently detected downstream of inputs of wastewater subject to advanced treatment ([Hicks et al., 2017](#); [Loomer et al., 2015](#)). Effects can be less consistent, however, for $\delta^{13}\text{C}$. In some systems, fish downstream of wastewater inputs exhibit consistent enrichment of $\delta^{13}\text{C}$ ([Hicks et al., 2017](#)) whereas in other systems no pattern has been observed.

In addition to effects mediated by wastewater-induced changes in $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$, concentrations of metals in fish can respond to effects of wastewater inputs on fish age, size, and growth rate (e.g., [Farkas et al., 2002](#)), which can all be influenced by wastewater effluent ([McMaster et al., 2005](#); [Tetreault et al., 2011](#)). Concentrations of bioaccumulating metals typically increase with fish size and age but decrease with faster growth rates and higher growth efficiency in a process referred to as somatic growth dilution (e.g., [Karimi et al., 2007](#); [Pickhardt et al., 2002](#); [Trudel and Rasmussen, 2006](#)). In colder, less productive Arctic environments, fish tend to grow more slowly and live longer than in temperate systems, which can result in relatively high levels of bioaccumulating contaminants ([Evans et al., 2005](#); [Schindler et al., 1995](#)). Eutrophication caused by wastewater inputs may induce somatic growth dilution of bioaccumulating metals in fish, but studies in the Arctic are lacking.

In this study, I sought to understand the state of a natural tundra wastewater wetland in Baker Lake, Nunavut, Canada, using data from both abiotic and biotic ecosystem components. Arctic grayling (*Thymallus arcticus*) and Burbot (*Lota lota*) were selected as sentinel species because they are widely distributed across the Arctic, highly valued by subsistence and recreational stakeholders, and differ in habitat use and trophic ecology. To investigate potential effects of municipal wastewater on downstream Arctic freshwater ecosystems, I: 1) characterized and compared the abiotic physicochemistry (i.e., water, sediment) of wastewater-affected sites (n=3 lakes) and reference sites (n=2 lakes); 2) evaluated differences in fish (Arctic grayling and Burbot) trophic ecology (i.e., $\delta^{15}\text{N}$, $\delta^{13}\text{C}$), energy storage (i.e., C:N, condition), and life history (i.e., growth) between wastewater-affected and reference sites; and, 3) investigated size-standardized differences in metal concentrations in fish muscle tissue between wastewater-

affected and reference sites. The approach I used advances the baseline understanding of the wastewater wetland, particularly in the context of the Hamlet's desire for upgrades to the treatment wetland. Additionally, the results could support the development of a community-based monitoring program as part of the municipal plan to enhance the wetland.

2.2 Methods

2.2.1 Study Area

Six lakes near the Hamlet of Baker Lake in the Kivalliq region of Nunavut (64.32°N, 96.02°W), were sampled in July and/or August of 2019 and 2021 ([Figure 2.1](#)). Water, sediment, bulk seston, Arctic grayling, and Burbot were collected. Three of the lakes comprise the natural tundra wastewater wetland north of the community (i.e., Lagoon Lake, Finger Lake, and Airplane Lake). Two reference lakes were selected based on accessibility and similar biogeochemistry to the largest lake in the wastewater wetland (i.e., Airplane Lake). Both reference lakes were located within 3 km of the Baker Lake wastewater wetland. Reference lake 1 (R1) is used for fly-fishing and Reference Lake 2 (R2) is commonly used for swimming by residents of Baker Lake (D. Nateela, pers. comm., Aug. 2021). The sixth lake is located immediately upstream (West) of the first wastewater lake in the lagoon-tundra treatment system (i.e., Upper Lagoon) and is not directly affected by wastewater. Data from Upper Lagoon Lake are visualized for comparison but are not included in the statistical analyses because the system is hydrologically connected to wastewater-affected lakes and thus this lake cannot be considered a reference system.

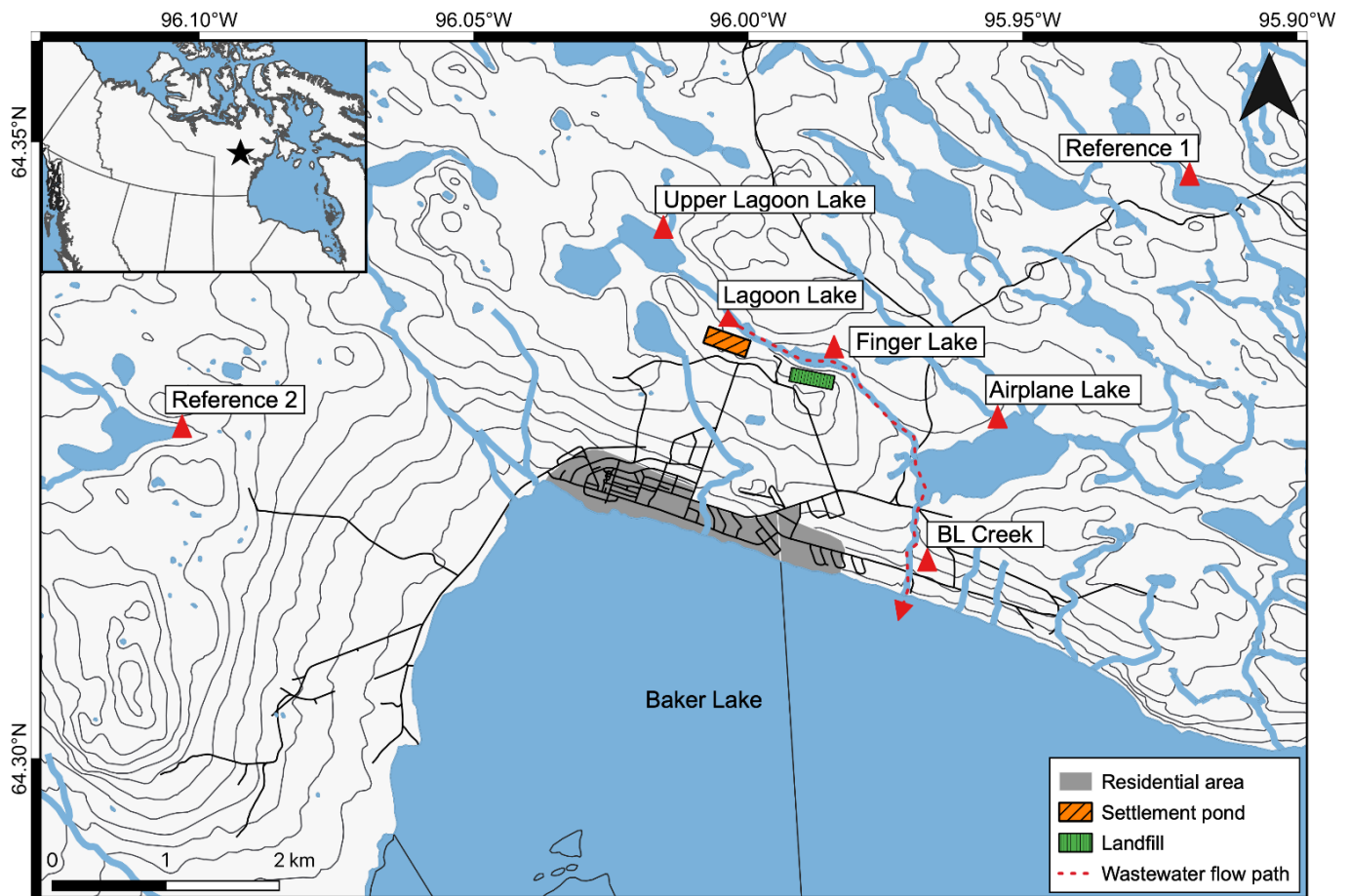


Figure 2.1 Map of the study sites in Baker Lake, Nunavut, Canada.

2.2.2 Sample Collection

Fish were collected using a backpack electrofisher (Smith-Root Inc., Vancouver), 3-hour sets of small-mesh gill nets (20 mm mesh size), 24-hour sets of fyke nets (18-26 mm mesh size), and angling in accordance with University of Waterloo Animal Care guidelines (AUP #41176). Arctic grayling were measured to the nearest millimeter for both fork and total length, while burbot were measured for total length only. Fish were then weighed to the nearest gram in the field and frozen immediately for shipment to the University of Waterloo. Catch data and biological data (means) are summarized in [Table S2.4](#).

To allow for quantification of among-system variability in $\delta^{15}\text{N}$ at the base of the food web (e.g., [Cabana and Rasmussen, 1996](#)), bulk seston was collected from each lake via horizontal tows approximately 30 cm below surface with an 80 μm mesh net.

Sediment and water were sampled in each of 2019 and 2021 in the approximate center of inflows and outflows in Finger Lake, Airplane Lake, R1, and R2. Lagoon Lake outflow and Baker Lake inflow were sampled in 2019 and 2021, respectively. A composite sediment sample was obtained by collecting the top 1 to 3 centimeters of sediment from three replicate Ekman dredge deployments. Water samples for total metal and nutrient analysis were collected by grab sampling, while samples for trace metals and major ions were collected using a peristaltic pump that pumped water through an acid-washed filter cartridge and muffled quartz QMA filter (2.2 μm pore size, (e.g., [U.S. EPA, 1996a](#)). In-field water quality parameters (i.e., temperature, pH, DO) were recorded at the time of sampling with a YSI handheld meter (ProDSS) and an Oxyguard Handy Polaris (Polaris 2). Near-surface (<30 cm) water samples were also collected in 1L amber bottles and filtered through ethanol-rinsed 0.42 μm Whatman GF/F filters for chlorophyll-*a* analysis. Individual filters were stored in opaque petri dishes, and filter volume was recorded.

Fish, bulk seston, and sediment samples were frozen at -20°C after collection. Water samples were kept at 4°C .

2.2.3 Sample preparation

Water samples were shipped to the University of Alberta Biogeochemical Service Laboratory, and frozen fish, sediment, and bulk seston were shipped to the University of Waterloo Fish Lab. Fish were thawed, and dorsal muscle tissue obtained from the left and right side of each fish was re-frozen in a 20 mL glass scintillation vial. An incision was made on the

ventral side of the fish, starting at the anus, and ending at the base of the gills. Once the internal organs were exposed, fish sex and maturity were determined visually. Gonads and livers were removed and weighed. Sagittal otoliths were removed by cutting through the skull of each fish and exposing the brain cavity. The membranous sac surrounding the otolith was removed before drying and storing in an envelope.

Sediment, bulk seston, and fish dorsal muscle tissue were freeze-dried using a LabConco FreeZone drier set at -54°C and 0.014 mBar for at least 48 hours. Dry samples were ground by motor and pestle until homogenized.

2.2.4 Laboratory analysis

Tissue samples, sediment, and water were analyzed at Biotron Center for Experimental Climate Change Research (University of Western Ontario, London, ON, CA), an ISO 17025 accredited laboratory. Total metal concentrations were determined using inductively coupled mass spectrometry following EPA Methods 3052 and 200.8 ([U.S. EPA, 1996b, 1994](#)). Metals selected for analysis (i.e., Al, Mn, Fe, Ni, Cu, Zn, As, Se, Sr, Cd, Hg, Pb) were determined from a literature review of metals frequently detected in municipal and landfill wastewaters ([Table S2.1](#)). Concentrations of total mercury (THg) were detected with a Milestone DMA-80 Direct Mercury Analyzer (Italy) DMA following EPA Method 1630 ([U.S. EPA, 2007](#)). Methyl mercury (MeHg) concentrations were determined using EPA Method 7473 for cold vapour atomic fluorescence spectroscopy ([U.S. EPA, 1998](#)) with a Tekran 2700 automated MeHg analysis system (Toronto, Canada). Sample quality assurance-quality control (QA/QC) was conducted in accordance with international standards ([Table S2.2](#)).

Dry fish tissue (weight 0.30-0.35 mg) and bulk seston (weight 0.40-0.45 mg) were weighed into 3.5 mm tin capsules and sent to the University of Waterloo Environmental Isotope Lab for analysis of stable carbon and nitrogen isotope ratios. Carbon and nitrogen stable isotope ratios expressed as parts per million (‰) delta values ($\delta^{13}\text{C}$, $\delta^{15}\text{N}$) were analyzed by a 4010 Elemental Analyzer (Costech Instruments, Italy) with a Delta Plus XL (Thermo-Finnigan, Germany) continuous flow isotope ratio mass spectrometer (CFIRMS). Accuracy was assured with international reference materials (IAEA-N1 & N2, IAEA-CH3 & CH6, USGS-40 & 41) and in-house reference materials (EIL-72, EIL-3, JSEC-01). Duplicate samples were analyzed every tenth sample to estimate precision. Stable isotope ratios for each sample were determined using the following formula:

Equation 1: $\delta^{13}\text{C}$ or $\delta^{15}\text{N} = [(\text{R}_{\text{sample}} - \text{R}_{\text{standard}})/\text{R}_{\text{standard}}] * 1000$, where $\text{R} = {}^{15}\text{N}/{}^{14}\text{N}$ or ${}^{13}\text{C}/{}^{12}\text{C}$.

Select inorganic water analyses were conducted at the ISO 17025 accredited Biogeochemical Analytical Service Laboratory (BASL, University of Alberta, Edmonton, AB, ON). Concentrations of total nitrogen (TN), total phosphorus (TP), ammonium (NH_4), and silica (SiO_2) were determined by flow injection analysis on a Lachat QuikChem 8500 FIA automated ion analyzer (USA). Chlorophyll-*a* was extracted with 95% ethanol from frozen filters for 12 hours before measurement with a Shimadzu RF-1501 Spectrofluorometer (North America Analytical & Measuring Instruments). Calcium (Ca) was measured using inductively coupled plasma – optical emission spectrometry on a Thermo Scientific ICAP6300 (USA), and chloride (Cl) was measured with ion chromatography on a Thermo Scientific Dionex DX600 (USA). Dissolved organic carbon (DOC) and sulfate (SO_4) were analysed at Western University. DOC was measured using phosphoric acid digestion followed by persulphate oxidation on an Aurora 1030W total organic

carbon analyzer (USA). SO_4 was measured with a Dionex ICS-1600 reagent free ion chromatography system (USA) and a AS-50 autosampler (USA).

2.2.5 Data analysis

2.2.5.1 Exploratory analysis and data pre-treatment

An initial exploratory data analysis was performed to check for data entry errors and allow for preliminary assessment of variables. Exploratory analyses included summarizing data, computing Pearson – product moment correlations, and performing simple linear regressions of well-established relationships (e.g., log-transformed fish length-at-weight).

Because previous research has shown that inputs of wastewater can affect $\delta^{15}\text{N}$ at the base of the food web, we first compared $\delta^{15}\text{N}$ ratios in a basal ecosystem component to evaluate if a baseline correction was necessary (Cabana and Rasmussen, 1996; Gartner et al., 2002). A *t*-test revealed that $\delta^{15}\text{N}$ ratios in bulk seston were significantly higher in wastewater-affected lakes than in reference lakes (*t*-test, $t(1,3) = 4.620$, $p = 0.019$). The elevated $\delta^{15}\text{N}$ in bulk seston in wastewater-affected lakes necessitated a baseline correction (e.g., Cabana and Rasmussen, 1994). Bulk seston was chosen to represent baseline $\delta^{15}\text{N}$ because it was the lowest trophic level biota captured in all lakes. Adjusted $\delta^{15}\text{N}$ values ($\delta^{15}\text{N}_{\text{adj}}$) were calculated as follows:

Equation 2: Baseline corrected $\delta^{15}\text{N}$ ($\delta^{15}\text{N}_{\text{adj}}$) = $\delta^{15}\text{N}_{\text{fish}} - \delta^{15}\text{N}_{\text{lake bulk seston}}$

Lipid content in fish tissue can introduce bias in $\delta^{13}\text{C}$ analysis due to the distinct isotopic composition of fats compared to proteins and carbohydrates (Focken and Becker, 1998). To account for this, a linear regression was conducted to establish the relationship between carbon isotope ratios of fish muscle tissue and the carbon-to-nitrogen ratio (C:N), which serves as a proxy for lipid content (Mintenbeck et al., 2008; Post et al., 2007). The relationship was not statistically

significant (*Linear regression*, $F(1, 90) = 1.290$, $p = 0.259$), and therefore, $\delta^{13}\text{C}$ values were left uncorrected (Post et al., 2007).

The length-weight relationship of fish may be represented by the following equation (Froese, 2006):

Equation 3: $W = aL^b$, where W is the weight of a fish (g), L is the length (cm), a is the intercept of the regression, and b is an exponential expression of the relationship between length and weight.

Arctic grayling exhibited isometric growth with proportional weight and length increase ($b=3$). Accordingly, Fulton's Condition Factor (K) was calculated (Froese, 2006; Ricker, 1975) to represent fish condition (Equation 4). In contrast, Burbot displayed negative allometry ($b < 3$), leading to the calculation of relative condition (K_n) for this species (Le Cren, 1951) (Equation 5).

Equation 4: Fulton's Condition Factor (K_{iso}) = $(L / W^3) * 100$, where L is length (mm), and W is total weight (g).

Equation 5: Relative Condition Factor = $K_n = w/W$, where w is the observed weight of a fish at a given length divided by W , the expected weight, which is calculated from the length-weight regression.

Insufficient sample size precluded growth curve modelling (e.g., Von Bertalanffy), and thus absolute growth rates were calculated. Absolute growth rate was calculated as follows:

Equation 6: Absolute growth rate, $Growth_{abs.}$ = $Length (mm) / Age (years)$

All statistical analyses were conducted in RStudio version 2023.12.1+402 using core packages from R version 4.3.3 (2024-02-29). Alpha was set to 0.05, unless otherwise indicated. When applicable, assumptions of homogeneity of variance were assessed by visually inspecting residual

plots. Data were $\log_{10}$ -transformed when necessary to meet assumptions of normality, which were assessed with Shapiro-Wilk statistics.

Linear Mixed-Effects Models (LMMs) were employed to address the structure of the dataset, in which lakes were nested within two site-types: wastewater-affected (n=3 lakes) and reference (n=2 lakes). In the linear mixed models employed in this thesis, site-type was a fixed effect, and lake was a random effect. Initially, linear mixed models were constructed to include both random intercepts and slopes, following the approach outlined by [Barr et al. \(2013\)](#). However, preliminary analysis revealed that the complexity of these models led to nonconvergence, likely due to the limited size of the dataset. Thus, models were simplified to include random intercept only, which is common for LMMs in ecology ([Harrison et al., 2018](#)). The random intercept formula can be represented as follows:

Equation 7: $\gamma_{ij} = \beta_0 + b_{0i} + \beta_1 X_{ij} + \epsilon_{ij}$

γ_{ij} , where is the response variable for the j^{th} observation in the i^{th} group (i.e., lake). β_0 is the overall intercept and b_{0i} is the random effect for the intercept associated with lake. β_1 is the fixed effect coefficient for the predictor variable (i.e., Site-type). X_{ij} represents the value of the predictor variable for each observation. Finally, ϵ_{ij} is the random error term associated with each observation, representing the unexplained variability once fixed and random effects were accounted for.

LMMs were constructed with 'lmer' in the package 'lme4' ([Bates et al., 2009](#)), and fit with restricted maximum likelihood (REML), providing unbiased estimates of variance and covariance parameters. If random effect variance estimates approached zero, and singularity was detected in the model, the model was deemed to be overparameterized. In such instances, the relationship

between the response variable and the fixed effect was analyzed using a linear model (LM) without a random effect (Zuur et al., 2009).

2.2.5.2 Characterization and comparison of abiotic physicochemistry between wastewater-affected and reference sites

Principal Component Analysis (PCA) was used to reduce dataset dimensionality and visualize differences in water chemistry across lakes. The PCA, performed with the ``prcomp`` function in R, included data from Upper Lagoon Lake for visualization purposes, though this lake was not included in the statistical analysis.

After conducting the PCA, a Bray-Curtis dissimilarity matrix was computed using the ``vegdist`` function from the ``vegan`` R package (Oksanen et al., 2007). This matrix quantifies beta diversity, which captures differences in water chemistry between wastewater-affected and reference sites. The matrix was analyzed with a Permutational Multivariate Analyses of Variance (PerMANOVA) using ``adonis2``, also from the ``vegan`` package. In this process, 1000 permutations were executed, and the statistical effect of site-type on water chemistry was assessed. Subsequently, individual Welch's *t*-tests were computed for each water quality parameter to identify significant differences between site-types. These methods were similarly applied to sediment chemistry parameters.

Additionally, metal concentrations in abiotic compartments were compared to the Canadian Council of Ministers of the Environment (CCME) guidelines for protection of aquatic life. In cases where CCME guidelines were not available, Federal Water Quality Guidelines (FEQGs) were employed (Health Canada, 2007).

2.2.5.3 Comparison of isotope ratios, absolute growth, C:N, and condition in fish between wastewater-affected and reference sites

To assess whether trophic level and carbon source in fish differed between wastewater-affected and reference site-types, $\delta^{15}\text{N}_{\text{adj}}$ and $\delta^{13}\text{C}$ ratios determined from Arctic grayling and Burbot muscle tissue were compared using linear mixed- models with fork length as a covariate. In this analysis, site-type (i.e., wastewater-affected vs. reference) was the fixed factor and lake was treated as a random factor. A similar approach was taken for absolute growth rate and C:N; for each species, absolute growth rate and C:N were compared between wastewater-affected and reference sites with a mixed model.

Prior to the comparison of fish condition between wastewater-affected and reference site-types, I tested for a potential effect of sex (fixed factor in a mixed model) on condition factor. There was no significant relationship between condition and sex for either Arctic grayling or Burbot ($\text{LMM}, \chi^2(1) > 0.025, p > 0.875$). Similar to above analyses, condition was then compared between wastewater-affected and reference sites with a mixed model; site-type was a fixed factor, and lake was a random factor.

2.2.5.4 Comparison of metal concentrations in fish tissue between wastewater-affected and reference sites

The current study compared metal concentrations in fish muscle tissues from wastewater-affected systems to those in reference systems. The analysis was limited to metals that exhibited significant differences in water or sediment between site-types. To adjust for variations in tissue metal concentrations attributable to fish body size, residuals were computed from linear relationships between fish length and metal content. These residuals were

subsequently added to the average fish metal concentration, thereby normalizing metal concentrations in relation to body size (Gotelli and Ellison, 2013). Size-adjusted metal concentrations were compared between wastewater-affected and reference site-types with a mixed model as described above. Arctic grayling and Burbot were analyzed independently.

Significant differences in tissue metal concentrations between fish in wastewater-affected and reference systems were further examined with Multiple Linear Regressions (MLRs). These MLRs explored how metal concentrations were related with factors such as trophic ecology (indicated by $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$), energy storage (evidenced by C:N ratio and condition), and growth rates. To ensure a robust analysis, multicollinearity was assessed using diagnostics from the 'olsrr' R package (Hebbali, 2020), such as variance inflation factor (VIF) and tolerance. The decision to include certain covariates in the model was informed by the extent of variance inflation, as determined by the eigenvalues (Morrison, 1990). Additionally, Akaike Information Criterion was employed to evaluate models and identify significant covariates model(s) (Akaike, 1969).

2.3 Results and Discussion

2.3.1 Comparison of abiotic physicochemistry between wastewater-affected and reference sites

2.3.1.1 Water Quality

Water quality differences between wastewater-affected (n=3) and reference (n=2) systems, as revealed by a principal component analysis (PCA), included concentrations of some metals and ions, DOC, and chlorophyll-*a*. The first principal component (Dim1; Figure 2.2) accounted for 62.4% of the variance; concentrations of manganese (Mn), iron (Fe), and copper (Cu) were mostly strongly associated with this first principal component. Correlations were also observed between Dim1, and all other metals analyzed except for calcium (Ca) and barium (Ba)

(see [Table S2.5](#)). The second principal component (Dim2) explained an additional 19.6% of variation, and this component was associated with concentrations of sulfate (SO_4^{2-}), dissolved organic carbon (DOC), and chlorophyll-*a* (chl*a*). As [Figure 2.2](#) illustrates, water chemistry in wastewater-affected lakes and reference lakes was distinct, with lakes closer to the wastewater input showing greater differences from reference lakes. These results are consistent with improved water quality along the Baker Lake wastewater flow path that was previously inferred from declines in concentrations of pharmaceuticals and PCBs ([Stroski et al., 2020](#)), wastewater-associated microbes ([Gromala et al., 2021](#)), and treatment performance parameters (e.g., carbonaceous oxygen demand, *E. coli*, TP) ([Yates et al., 2012](#)), and suggests that the lagoon tundra wastewater wetland does result in some improvements in water quality downstream of influent. Despite this, all wastewater-affected lakes exhibit higher metal concentrations and greater productivity than reference lakes ([Figure 2.3](#)). Significant differences were observed between reference and wastewater-affected lakes for both concentrations of metals (including Mn, Fe, Cu, Zn, Ba, and Pb (*PerMANOVA*, $F(1, 8) = 17.319$, $p=0.007$) and productivity-related parameters, including chlorophyll-*a*, total nitrogen (TN), and total phosphorus (TP) (*PerMANOVA*, $F(1, 8) = 5.947$, $p=0.035$).

Identifying and evaluating individual water quality parameters that are significantly affected by wastewater is crucial for effective monitoring and decision-making regarding future upgrades in water treatment. Individual *t*-tests ([Table 2.1](#)) revealed significantly higher concentrations of Mn, Fe, Cu, and Pb in wastewater-affected lakes than in reference lakes. Higher concentrations of chlorophyll-*a*, TN, and TP were also significantly different in wastewater-affected sites. These parameters, along with Secchi depth, can be used to determine the Trophic

State Index (TSI) of a lake (Carlson, 1977). Using this index, Airplane Lake and Finger Lake are classified as eutrophic based on elevated productivity parameters (detailed in Table 2.2), while lakes R1 and R2 are categorized as oligotrophic, typical of low-nutrient, Arctic waterbodies (Huser et al., 2022). Understanding the trophic state of water bodies affected by human activities can guide the monitoring and management of Arctic ecosystems (EMAN-North, 2005).

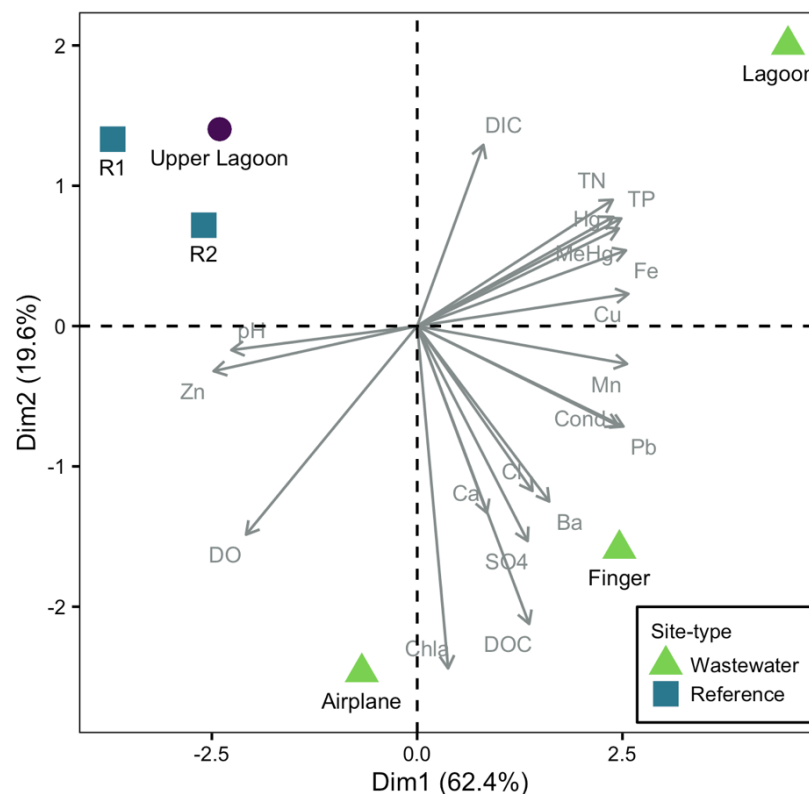


Figure 2.2 Principal component analysis (PCA) biplot of water quality parameters in wastewater- affected (triangle) and reference (square) lakes. Individual lakes are labeled on the plot. Results for Upper Lagoon Lake (UL; Circle) are included for information only and were not used to determine eigenvalues. Dimension 1 (Dim1) of the PCA explains 62.4% of the variation among lakes and is highly correlated with several analysed metals. Dimension 2 (Dim2) explains an additional 19.6% of variation among lakes and is highly correlated with parameters that indicate primary productivity, including Chla, DOC, and SO₄²⁻, which are responsible for 24.3%, 19.6%, and 10.0% of Dim2 variation, respectively. Lakes closest to wastewater input (i.e., Lagoon and Finger Lake) were separated from reference lakes across Dim1, indicating high contributions of metal concentrations contribute to dissimilarity. The furthest downstream wastewater lake (i.e., Airplane Lake) separates from reference lakes on Dim2 more than Dim1, indicating that metal concentrations are more similar to reference sites in this furthest downstream lake.

Table 2.1 Comparison of water chemistry parameters between wastewater-affected and reference sites using Welch's *t*-tests, with false discovery rate adjustments for multiple comparisons. In wastewater-affected sites, concentrations of Mn, Fe, Cu, and Pb were significantly greater than reference sites. Concentrations of Zn, Ba, and MeHg were not significantly different between site-types, although MeHg was limited to one year of data due to sample contamination. Lake productivity parameters (i.e., chl_a, TN, and TP), as well as conductivity were significantly higher in wastewater-affected sites. There was no significant difference in dissolved oxygen (DO) concentrations between site-types, though low DO concentrations were noted in Lagoon Lake. When compared against the Canadian Council of Ministers of the Environment (CCME) guidelines and Federal Water Quality Guidelines (FEQG) for the protection of freshwater aquatic organisms, no exceedances were observed for dissolved metal concentrations. However, DO levels in Lagoon Lake were below acceptable CCME guidelines for all life stages.

Parameter	df	t	p	Guideline
Manganese (Mn)	1, 5.021	3.892	<u>0.020</u>	<CCME
Iron (Fe)	1, 5.034	3.412	<u>0.022</u>	<FEQG
Copper (Cu)	1, 5.121	3.995	<u>0.020</u>	<FEQG
Zinc (Zn)	1, 3.848	-0.136	0.551	<CCME
Barium (Ba)	1, 7.245	0.0204	0.492	No guidelines
Lead (Pb)	1, 6.276	2.789	<u>0.026</u>	<FEQG
Methylmercury (MeHg)	1, 2.083	1.874	0.138	<CCME
Chlorophyll- <i>a</i> (chl _a)	1, 5.005	2.432	<u>0.025</u>	Site specific
Total nitrogen (TN)	1, 5.001	2.897	<u>0.025</u>	<CCME
Total phosphorous (TP)	1, 5.005	3.007	<u>0.030</u>	Site specific
Dissolved oxygen (DO)	1, 4.152	-1.027	0.820	>CCME (Lagoon Lake)
Conductivity (Cond)	1, 4.459	4.952	<u>0.003</u>	No guidelines
Sulfate (SO ₄)	1, 4.016	2.661	<u>0.028</u>	No guidelines

Lagoon Lake, the first lake in the treatment wetland, offers a unique opportunity to study the more severe impacts of wastewater on Arctic freshwater ecosystems. Lagoon Lake is classified as hypereutrophic (Table 2.2), which sets it apart from both reference lakes and other wastewater-affected lakes. Elevated metal concentrations in Lagoon Lake, which is typical of municipal wastewater (e.g., Chipasa, 2003; summarized in Table S2.1), separate it on Dim1 (Figure 2.2). On Dim2, Lagoon Lake differentiates from Airplane Lake and Finger Lake due to its notably low concentration of chlorophyll-*a*, which accounts for the greatest variance (24.3%) of

any single dimension. Low chlorophyll-*a* concentrations in Lagoon Lake are likely a result of high turbidity and short residence times (observed in [Figure 2.4](#)), which are factors that may also contribute to the extremely low dissolved oxygen concentrations ([Wetzel, 2001](#)). As depicted in [Figure 2.3](#), the dissolved oxygen levels in Lagoon Lake were notably lower than those observed in other study lakes. While DO concentrations between wastewater-affected and reference sites were not significantly different ([Table 2.1](#)), mean DO in Lagoon Lake was only 3.32 mg/L, which is substantially below the minimum 9.5 mg/L required for the early life stages of cold freshwater organisms such as Arctic grayling, and also below the 6.5 mg/L necessary for older life stages ([CCME, 2001](#)). This insufficiency was evident when six young-of-year Arctic grayling were found dead or in poor condition after a minnow trap set of fewer than three hours during the 2019 field season. Reduced oxygen levels are known to cause lethal and sublethal effects in various aquatic organisms, especially fish (e.g., [Pollock et al., 2007](#)).

Low concentrations of DO in lagoon-tundra wastewater wetlands can impede treatment processes as well as pose risks for aquatic life. These wetlands rely on either aerobic or anaerobic microbial activities, or a combination of both, to transform contaminants ([Chouinard et al., 2014](#); [Yates, 2012](#)). Aerobic activity, which uses oxygen to convert organic wastes into biomass (i.e., microbial cells) and carbon dioxide, is typically more effective than anaerobic processes ([Chan et al., 2009](#)). When available DO fails to meet the oxygen requirements of aerobic bacteria, there is a shift towards anaerobic processes, which can result in a slower treatment performance and lower quality effluent due to incomplete decomposition of organic matter ([Chan et al., 2009](#)). Residents of Baker Lake have reported increased odors from the local wastewater wetland in recent years. This rise in odor, typically a by-product of anaerobic activity, correlates with findings

by Ragush et al., (2015) who found that odors, predominantly a by-product of anaerobic activity, were noted in years when anaerobic processes dominated in three Arctic wastewater stabilization ponds. However, detailed data collection is needed to clarify biogeochemical processes in Lagoon Lake.

Table 2.2 The 2019 and 2020 average Trophic State Indices (TSI) calculated for each lake from three index variables: chlorophyll-*a* (chl_a), total phosphorous (TP), and Secchi depth (SD). Equations are as follows: $TSI_{(chl_a)} = 9.81 \ln(chl_a) + 30.6$, $TSI_{(TP)} = 14.42 \ln(TP) + 4.15$, and $TSI_{(SD)} = 60 - 14.41 \ln(SD)$. TSI Classification: oligotrophic (O): mesotrophic (M), eutrophic (E), hypereutrophic (HE). Values are means ($\pm$ SE).

Parameter	Upper Lagoon	Lagoon	Finger	Airplane	R1	R2
Chlorophyll- <i>a</i> (µg/L)	0.96 ±0.32	2.18 ±0.31	13.00 ±5.40	30.60 ±7.30	0.60 ±0.05	0.67 ±0.20
TP (µg/L)	2.00 ±1.83	683.50 ±113.49	331.25 ±25.75	74.00 ±9.32	2.75 ±2.06	2.50 ±1.08
Secchi Depth (m)	3.48 ±0.42	0.10 ±0.00	0.78 ±0.21	0.53 ±0.09	4.20 ±0.73	4.95 0.35
Trophic State Index	28.79	76.57	69.06	66.51	27.88	27.00
TSI Classification	O	HE	E	E	O	O

To assess the risk of elevated metal concentrations in the lagoon-tundra wastewater wetland, this study applied guidelines from the Canadian Council of Ministers of the Environment (CCME) and the Federal Water Quality Guidelines (FWQG) to protect freshwater organisms (Table 2.1). The CCME provides flexible, regionally adaptable guidelines, whereas the FWQG offers specific federal guidance. While no dissolved metals exceeded guideline limits, the result for Cu exemplifies the need for ongoing monitoring in areas affected by human activity. The FWQG for Cu is dynamic and relies on the Biotic Ligand Model to integrate site-specific water chemistry that affects bioavailability and toxicity (Di Toro et al., 2001; ECCC, 2021; Santore et al., 2001). Factors such as pH, dissolved organic carbon, and major ion concentrations (i.e., calcium, magnesium,

sodium, and potassium) affect toxicity thresholds (Playle et al., 1992). Importantly, Nunavut records higher surface water Cu concentrations than many other provinces and territories (ECCC, 2021), which should also be considered in management decisions. Despite the lack of immediate

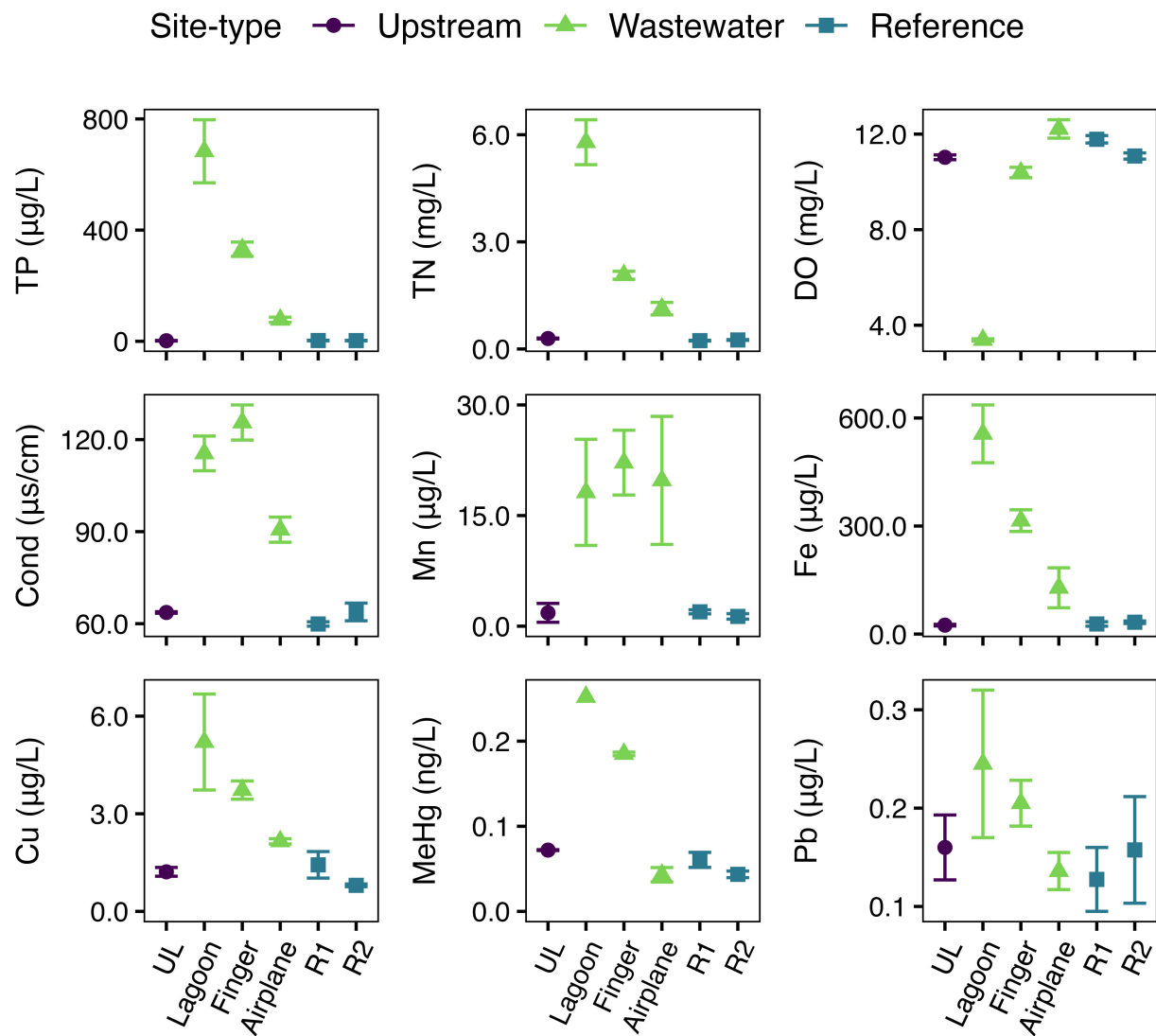


Figure 2.3 Arithmetic mean ($\pm$ SE) values for water quality parameters, using all available data (2019 and 2021 and all technical replicates). Note that for MeHg only data from 2021 were available. Wastewater-affected sites (i.e., Lagoon Lake, Finger Lake, and Airplane Lake) had higher mean values compared to reference sites (i.e., R1 and R2) for all parameters except DO. DO in Lagoon Lake was below the lowest acceptable concentration for protection of aquatic life in Lagoon Lake (CCME). Upper Lagoon Lake (UL) is directly upstream of the wastewater wetland and is included for visual comparison only.

toxicity risks, continued monitoring of the Baker Lake lagoon-tundra wastewater wetland is essential to capture seasonal and inter-annual fluctuations in metals and other contaminants, particularly in the context of assessment after wastewater treatment upgrades.

The Nation Standard of Canada (CSA W203:19) for wastewater treatment in the Arctic has only recently recognized wetlands as essential to Northern wastewater management, previously considered only as part of the receiving environment. This shift highlights the need for broader research into treatment processes throughout the entire wetland, focusing not just on points of influent and effluent. Comprehensive data collection and modeling, as suggested by [Balch et al., \(2018\)](#) are crucial for these investigations, particularly in understanding seasonal and annual variations.

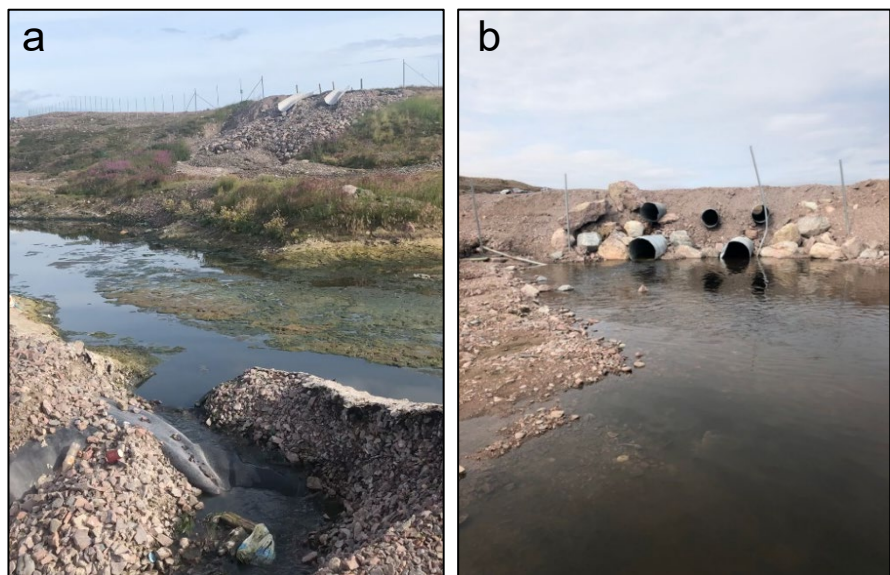


Figure 2.4 Photographs from 2019 field work in Baker Lake, NU showing: a) a settlement pond adjacent to the wastewater wetland, with a visibly damaged gravel berm that allows effluent to bypass directly into the wetland, and b) the inflow of effluent from Lagoon Lake to Finger Lake, facing west. The water is notably turbid and large amounts of periphyton are visible on submerged boulders.

2.3.2.2 Sediment quality

In the Principal Component Analysis (PCA) of sediment chemistry parameters, the first two dimensions captured a significant portion of total variation ([Figure 2.5](#)). The first dimension (Dim1) accounted for 46.8% of the variation and was primarily influenced by concentrations of

manganese (Mn), total mercury (THg), and iron (Fe). The second dimension (Dim2) explained an additional 32.3% of the variation and was associated with concentrations of zinc (Zn), copper (Cu), and methylmercury (MeHg). In a pattern similar to that observed for water quality ([Figure 2.2](#)), lakes nearer to the influent source, such as Lagoon Lake and Finger Lake, exhibited more distinct sediment chemistry compared to Airplane Lake, the most downstream wastewater lake, and reference lakes. The spatial positioning of lakes in the PCA, along with their correlations to specific metals, helps discern the effects of wastewater on the chemical, physical, and biological characteristics of lake sediments in this system.

Lagoon Lake was characterized by a negative score on the first dimension (Dim1), and a positive score on the second dimension (Dim2), which reflected higher concentrations of metals, such as MeHg, that scored positively on Dim2. It is not surprising that MeHg is high in sediments of Lagoon Lake, given its hypereutrophic state and low dissolved oxygen concentrations, conditions known to promote biological methylation of mercury. The contrasting concentrations of MeHg in the sediment of Lagoon Lake and Finger Lake are interesting. Lagoon Lake sediment has exceptionally high MeHg levels, nearly eight times higher than those found in reference sites ([Figure 2.6](#)). Furthermore, the proportion of Hg that is methylated in Lagoon Lake is higher than in Finger Lake: 9.1% compared to only 1.7%, respectively (see [Table S2.3](#) and [Figure 2.6](#)). Both lakes have abundant energy sources for bacteria that could methylate mercury, as evidenced by high loss-on-ignition values for sediments (see [Table S2.3](#)), but as discussed above, Lagoon Lake has very low dissolved oxygen concentrations. In anaerobic conditions, such as those observed in Lagoon Lake, bacteria can convert inorganic mercury to the more toxic MeHg using sulfate (SO_4^{2-}), carbon dioxide, hydrogen, or iron as electron acceptors ([Paranjape and Hall, 2017](#)).

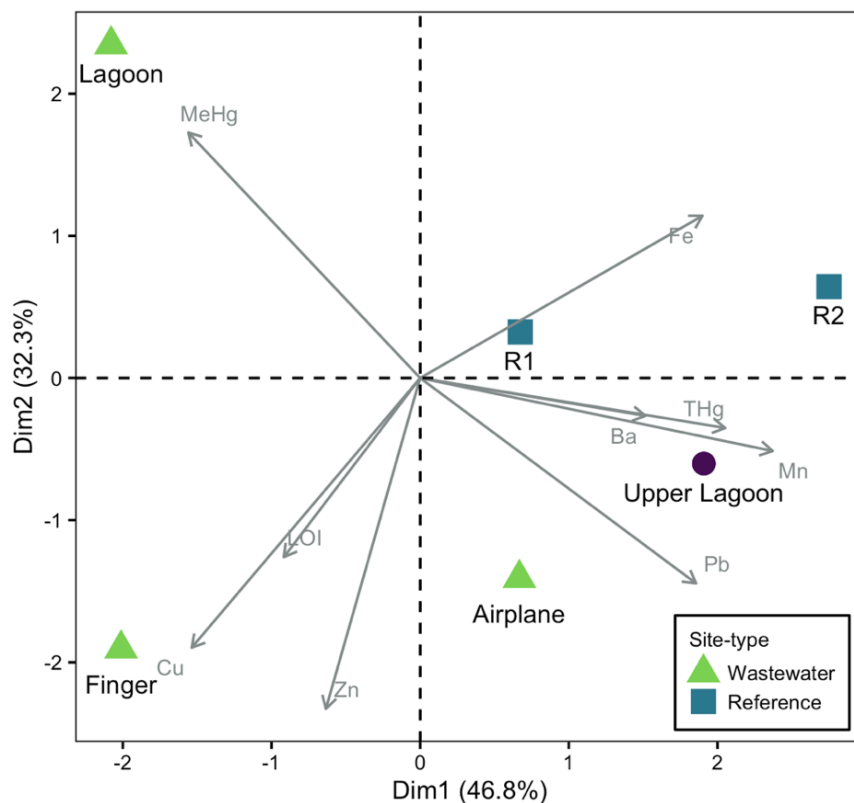


Figure 2.5 Principal component analysis (PCA) biplot of sediment parameters in wastewater-affected (triangle) and reference (square) sites. Individual lakes are labeled on the plot, including Upper Lagoon Lake (UL; Circle) which is provided for information only and was not used to estimate eigenvalues. Dimension 1 (Dim1) of the PCA explains 46.8% of the variation among lakes and is highly correlated with concentrations of Mn, THg, and Fe. Dimension 2 (Dim2) explains an additional 32.3% of variation among lakes and is highly correlated with concentrations of Zn, Cu, and MeHg. Lakes closest to the wastewater input (i.e., Lagoon and Finger Lake) differ from reference lakes across both dimensions, as influenced by [MeHg] and [Cu], respectively. The final wastewater Lake (i.e., Airplane Lake) and R1 have similar positions on Dim1, therefore, variation may be explained by parameters strongly correlated with Dim2 (Zn, Cu, MeHg). Airplane Lake differs from R2 across both dimensions.

Distinguished by its negative positioning on both dimensions in the PCA, Finger Lake diverges from Lagoon Lake on Dim2 and is strongly associated with Cu and Zn. This pattern implies there are environmental influences shared with Lagoon Lake on Dim1, likely due to the downstream flow of wastewater. However, the divergence of Finger Lake on Dim2 indicates that sediment chemistry might be affected by additional sources of metals, such as the landfill on the

lake's south shore (Figure 2.1). Therefore, sediment chemistry in Finger Lake appears to be influenced by municipal wastewater and direct additions of Cu and Zn from landfill leachate.

Further downstream, the position of Airplane Lake in the sediment chemistry PCA indicates a moderate impact of wastewater (Figure 2.5). The less extreme but positive loading on Dim1 suggests a diluted impact of metals from upstream contamination sources. Moreover, the negative Dim2 loading aligns with a reduced influence of the factors associated with higher MeHg concentrations, similar to the pattern observed in Finger Lake. The change in metal profile could be due to ongoing environmental processing such as dilution, biological uptake, or chemical transformations that occur throughout the wastewater treatment wetland. Managing the Baker Lake wastewater wetland may require lake-specific strategies. For instance, decreasing nutrient additions to Lagoon Lake could enhance treatment success, whereas Finger Lake might require interventions that addresses the impacts of both municipal wastewater and landfill run-off.

Overall, there was no significant difference in sediment chemistry between the wastewater-affected sites and the reference sites (*PerMANOVA*, $F(1, 7) = 1.592, p = 0.225$). However, consistent with earlier PCA observations, significant differences were found between site-types in the levels of Copper (Cu) and Zinc (Zn). Cu concentrations were significantly higher in wastewater-affected sites compared to reference sites (*t-test*, $t(1, 5.338) = 5.660, p = 0.014$), as were Zn concentrations (*t-test*, $t(1, 4.982) = 3.874, p = 0.041$). In contrast, no significant differences were observed in the concentrations of Mn, MeHg, Iron, Fe, and Pb between the two types of sites, as detailed in (Table 2.3).

These results align with previous research on constructed wetlands used for municipal wastewater treatment in temperate climates. Specifically, Lesage et al. (2007a, 2007b) and the

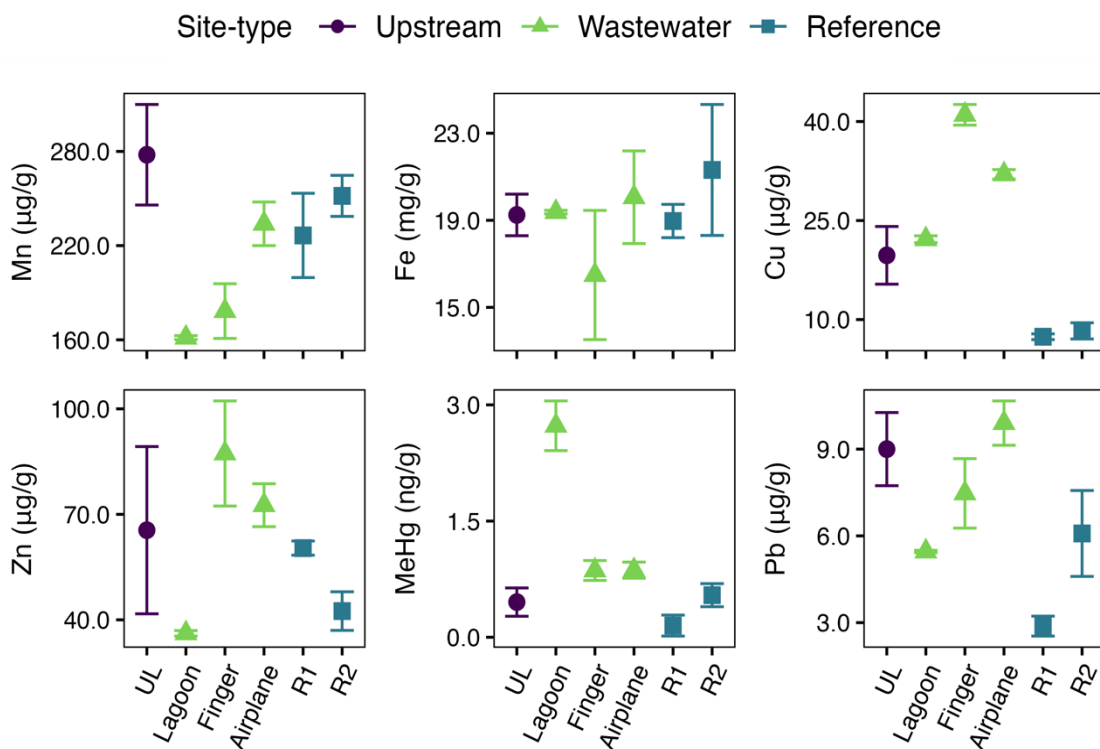


Figure 2.6 Arithmetic mean ($\pm$ SE) values for sediment metal concentrations, using all available data (2019 and 2021 and all technical replicates). A distinctly high concentration of MeHg was measured in Lagoon Lake compared to other impacted lakes and non-impacted lakes. THg measured in all study lakes was well below the CCME freshwater guideline (PEL: 0.486 $\mu\text{g/g}$ dry weight) for the protection of aquatic life.

and Vymazal and Krása (2003), who found significant accumulation of Cu and Zn in early treatment zones of temperate constructed wastewater wetlands, but not of Pb, Fe, and Mn. There is a distinct scarcity of data for natural (i.e., non-engineered) wastewater wetlands in polar regions, making direct comparisons with these environments difficult. This knowledge gap emphasizes the need for more investigations of metal accumulation in the sediment of Arctic wastewater wetlands, with a focus on understanding biotic and abiotic factors that drive complex interactions and spatio-temporal variability.

In general, wastewater treatment wetlands are recognized as effective sinks for metals, due to conditions that promote retention, such as high organic matter content and low oxygen

levels (Wetzel, 2001). Organic matter plays a crucial role not only in forming complexes with metals like Cu and Zn but also in reducing the redox potential, which further influences the precipitation and adsorption of metals. Cu tends to form strong complexes with ligands found in algae (Sunda and Lewis, 1978), while Zn forms complexes of varying strength, depending on the chemical conditions (Xue et al., 1995). Complexation with organic matter, coupled with microbial activity, significantly alters the solubility and bioavailability of these metals. As organic material undergoes denitrification, metals become incorporated into the sediment layers. The enrichment of [Cu] and [Zn] in wastewater-affected sediments is further enhanced by their strong adsorption to Fe(III) oxides and hydroxides, which act to immobilize and retain these metals within the sediment (Dunbabin and Bowmer, 1992; Xue et al., 1995). Despite expectations, mean [Fe] was lower in wastewater-affected sites compared to reference sites, although not significantly (*t*-test, $p>0.05$) (Figure 2.6). This result can be attributed to biogeochemical interactions in wetlands, including adsorption, precipitation, and microbial activity that are heavily influenced by oxygen availability (Fredrickson and Gorby, 1996). Wastewater wetland sediments typically exhibit anaerobic conditions due to decomposition of organic matter and the high oxygen demand from microorganisms (Wetzel, 2001). In anaerobic environments, iron-reducing bacteria use Fe compounds as electron acceptors, converting less soluble Fe(III) to more soluble Fe(II) (Fredrickson and Gorby, 1996; Weber et al., 2006). This conversion could explain the higher Fe concentrations observed in water (Figure 2.3) but not sediment (Figure 2.6) within the Baker Lake wastewater wetland. The biogeochemical role of Fe in wastewater treatment wetlands is reviewed in-depth by Wu et al. (2019), with specific focus on retention and remobilization.

Table 2.3 Comparison of sediment chemistry between wastewater-affected and reference sites using Welch's t-tests with corrections for false detection rates. Significant differences in copper and zinc concentrations were found. Mean sediment concentrations were compared to guidelines set by the Canadian Council of Ministers of the Environment (CCME) when available. Copper concentrations in Lagoon Lake were between the threshold effect level (TEL) and probable effect (PEL) level, signifying that negative effects on biota occasionally occur at similar concentrations.

Parameter	<i>df</i>	<i>t</i>	<i>p</i>	Guideline
Copper (Cu)	1, 5.338	5.660	<u>0.014</u>	CCME (>TEL, <PEL) in Finger Lake
Manganese (Mn)	1, 4.876	0.404	0.704	No guideline
Zinc (Zn)	1, 4.982	3.874	<u>0.041</u>	CCME (<PEL)
Lead (Pb)	1, 5.911	2.158	0.139	CCME (<PEL)
Methylmercury (MeHg)	1, 6.917	1.247	0.380	No guideline
Iron (Fe)	1, 6.007	1.211	0.380	No guideline
Barium (Ba)	1, 3.976	1.115	0.382	No guideline
Loss on Ignition (LOI)	1, 1.513	2.417	0.176	No guideline

To conclude the examination of metal dynamics in the wastewater wetland sediments, concentrations were compared to the Canadian Council of Ministers of the Environment guidelines for the protection of aquatic life (CCME, 2001), as outlined in (Table 2.3). The concentration of total mercury (THg) in all surveyed lakes was below the CCME's Probable Effect Level (PEL) of 0.486 µg/g dry weight. Although the CCME guidelines include THg, they exclude MeHg due to limited data. Typically, MeHg is found to constitute only 0.01% to 10% of the THg in sediments (CCME, 2001). Our study found that MeHg comprised 9.1% of the THg in Lagoon Lake, placing it at the high end of this range, while all other lakes in the study recorded proportions below 2%. This finding indicates that the biota in the Baker Lake lagoon-tundra wastewater wetland may be exposed to higher levels of MeHg in some systems, potentially posing risks to health and survival (Cabana et al., 1994; Watras et al., 1998). Furthermore, the

study observed that sediment copper (Cu) concentrations in Finger Lake fell within the range between the CCME's Threshold Effect Level (TEL) and PEL. This suggests the possibility of intermittent adverse effects on the biota.

2.3.2 Comparison of isotope ratios, absolute growth, C:N, and condition in fish between wastewater-affected and reference sites

To evaluate the broader implications of municipal wastewater on Arctic tundra wetland ecosystems, this research included evaluation of both biotic and abiotic parameters. The biotic component of the investigation focused on two large-bodied fish species, Arctic grayling (*Thymallus arcticus*) and Burbot (*Lota lota*). Indicators of trophic ecology (i.e., $\delta^{15}\text{N}$, $\delta^{13}\text{C}$), energy storage (i.e., C:N, condition), and absolute growth were compared between wastewater-affected and reference sites. The previously identified differences in water and sediment quality between site-types provided a compelling context for understanding how these fish, with their unique feeding strategies and environmental sensitivities, responded to differences in abiotic conditions. The study of isotopic ratios in fish muscle tissue can provide preliminary insights into fish foraging behaviour and trophic position (Cabana and Rasmussen, 1994; France, 1995). This is particularly relevant when assessing the impact of nutrient additions and increased primary productivity on fish ecology (Kidd et al., 2012; Peterson and Fry, 1987; Pickhardt et al., 2002). Interestingly, the isotopic ratios in Burbot were similar between reference and wastewater-affected site-types (Figure 2.7). The lack of significant differences in [length-adjusted] $\delta^{15}\text{N}_{\text{adj}}$ and $\delta^{13}\text{C}$ in muscle tissue between site-types, detailed in Table 2.5, suggests minimal influence of wastewater on trophic position or foraging behaviour in Burbot. Conversely, length-adjusted $\delta^{13}\text{C}$ in Arctic grayling muscle tissue was significantly enriched in wastewater-affected sites compared to

reference sites ($LM, F(1, 60) = 9.475, p = 0.003$). Similar to Burbot, Arctic grayling muscle tissue $\delta^{15}N_{adj}$ did not significantly differ between site-types (Table 2.4). This finding implies that while the introduction of wastewater into lakes may alter the carbon sources (i.e., energy pathways) of Arctic grayling, it does not significantly affect their trophic position. For a definitive understanding of the isotope ecology of fish in systems affected by wastewater near Baker Lake, more comprehensive models, such as a two-member mixing model, are necessary; this would require additional sampling efforts.

Hayden et al. (2019) used two-member mixing models to assess the response of subarctic lake ecosystems to eutrophication from land use and climate change. Their study identified a significant shift from benthic to pelagic resource use, evidenced by depleted $\delta^{13}C$ values in biota

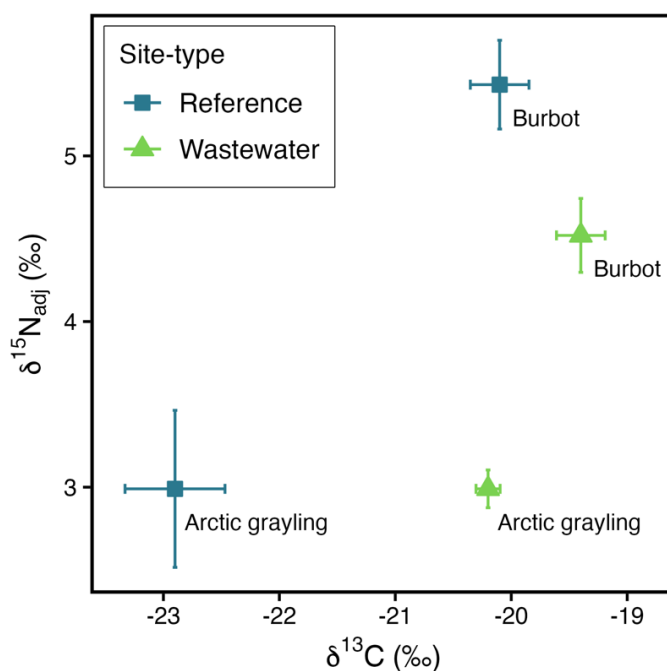


Figure 2.7 Isotope biplot of length-adjusted mean $\pm$ standard error $\delta^{15}N_{adj}$ and $\delta^{13}C$ for Arctic grayling and Burbot from wastewater-affected sites ($n=3$; green triangles) and reference sites ($n=2$; blue squares) near Baker Lake, Nunavut, Canada. A significant difference in Arctic grayling $\delta^{13}C$ was observed ($LM, F(1, 60) = 9.475, p = 0.003$), but not in $\delta^{15}N_{adj}$ or either isotope for Burbot. $\delta^{15}N_{adj}$ values are mathematically corrected for baseline values in each lake.

across gradients of increased productivity and temperature. In contrast, Arctic grayling from wastewater-affected sites in Baker Lake were $\delta^{13}\text{C}$ -enriched compared to reference sites, with measurements of -20‰ and -23‰, respectively, aligning with similar findings downstream from municipal wastewater plants in temperate watersheds ([Brown et al., 2011](#); [Hicks et al., 2017](#); [Robinson et al., 2016](#)). Understanding energy pathways, particularly benthic-pelagic coupling, is crucial as carbon source can significantly impact fish growth rates ([Moslemi-Aqdam et al., 2021](#)), and ultimately metal bioaccumulation in subarctic lakes ([Moslemi-Aqdam et al., 2022](#)). Such insights are increasingly important considering the complex interactions that shape carbon pathways in human-altered landscapes, including wastewater inputs, climate change, landfill leaching, and increased impervious surfaces (e.g., [Hayden et al., 2019](#)).

Investigators have struggled to explain enriched $\delta^{13}\text{C}$ observed in organisms from environments affected by wastewater ([Hicks et al., 2017](#); [Wayland and Hobson, 2001](#)). [Wayland and Hobson \(2001\)](#) suggest that enrichment could stem from rapid autotrophic growth in sewage-impacted systems, fostering bottom-up enrichment of $\delta^{13}\text{C}$ through the food web. This hypothesis is supported by observed $\delta^{13}\text{C}$ enrichment in Arctic grayling muscle tissue from the Baker Lake wastewater wetland compared to reference systems ([Figure 2.7](#)); Bulk seston samples, which represent the base of the pelagic food web, and periphyton samples, which represent the base of the benthic food web, were also enriched in wastewater-affected systems relative to reference systems, although sample sizes were extremely limited ([Table S2.5](#)). The pattern of $\delta^{13}\text{C}$ enrichment in wastewater-affected systems was not reflected in Burbot, as there was no significant difference in $\delta^{13}\text{C}$ between individuals from wastewater-affected and reference systems ([Table 2.5](#)). Burbot are a bottom-feeding fish ([Scott and Crossman, 1973](#)), and

we did not fully characterize isotope ecology of the profundal habitats where Burbot often forage. It is possible that the effects of nutrient enrichment vary among pelagic, littoral benthic, and profundal habitats, as well as among different basal food sources. This variability warrants further investigation, ideally across both temperate and subarctic ecosystems.

Long-term studies in the Grand River Watershed, Ontario, Canada, revealed significant $\delta^{13}\text{C}$ enrichment in Rainbow Darters (*Etheostoma caeruleum*) downstream of wastewater treatment plants, compared to upstream populations (Hicks et al., 2017). The observed enrichment in Rainbow Darters could be attributed to their generalist feeding strategy, which enables them to exploit the more nutrient-dense benthic zones (Brown et al., 2011; Robinson et al., 2016). Similarly, Arctic grayling are also generalists but primarily feed at the surface or in pelagic zones (Scott and Crossman, 1973). Further research is needed to determine if Arctic grayling in wastewater-impacted systems become increasingly reliant on benthic-derived carbon, which could explain the observed $\delta^{13}\text{C}$ enrichment. Their nature as visual predators suggests that this enrichment could be due to impaired visibility in open water, reduced pelagic foraging, and a compensatory increase in benthic feeding (Stewart et al., 2007b). Alternatively, the enriched $\delta^{13}\text{C}$ in Arctic grayling from these systems might simply reflect enriched $\delta^{13}\text{C}$ in bulk seston and periphyton. Future studies should include isotope analyses on more spatial and temporal replicates of basal organisms across multiple habitats. Understanding how sewage-derived nutrients influence the isotopic ecology of fish and their assimilation in the food web will provide valuable insights into contaminant exposures in aquatic ecosystems (Aguilar-Betancourt et al., 2016; deBruyn and Rasmussen, 2002; Hicks et al., 2017).

Table 2.4 Summary of results from Linear Models (*LM*) and Linear Mixed Models (*LMM*) assessing differences between wastewater-affected and reference sites in trophic ecology (i.e., $\delta^{15}\text{N}$, $\delta^{13}\text{C}$), energy storage (i.e., C:N, condition), and growth of Arctic grayling. The random effect of lake was tested for each covariate. If variance estimates approached zero, and singularity was detected, the model was deemed to be overparameterized. In such instances, relationships were analyzed using a linear model (*LM*) without a random effect. Parameter estimates (β), standard errors (SE), degrees freedom (df), the test statistic (i.e., χ^2 ; chi-square or F; F value), and significance (p) are shown. Significant differences ($\alpha = 0.05$) are underlined and bold.

Arctic grayling Model					Results		
Covariate	Model Type	Fixed Effect	β	SE	df	Test Statistic	p
$\delta^{15}\text{N}_{\text{adj}}$	<i>LMM</i>	Site-type	13.626	7.796	1	$\chi^2 = 3.054$	0.081
		Length	4.410	3.086	1	$\chi^2 = 2.024$	0.153
		Site-type*Length	-5.846	3.135	1	$\chi^2 = 3.478$	0.062
$\delta^{13}\text{C}$	<i>LM</i>	Site-type	-0.130	0.033	1, 60	F = 15.471	<u><0.001</u>
		Length	<0.001	<0.001	1, 60	F = 7.216	<u>0.009</u>
		Site-type*Length	<0.001	<0.001	1, 60	F = 9.475	<u>0.003</u>
Growth _{abs}	<i>LMM</i>	Site-type	130.111	42.402	1	$\chi^2 = 4.845$	<u>0.028</u>
		Length	0.1483	0.126	1	$\chi^2 = 1.523$	0.966
		Site-type*Length	-0.302	0.129	1	$\chi^2 = 4.046$	<u>0.044</u>
C:N	<i>LM</i>	Site-type	-2.418	1.510	1, 60	F = 2.509	0.118
		Length	-1.097	-0.607	1, 60	F = 3.267	0.076
		Site-type*Length	1.083	0.615	1, 60	F = 3.103	0.083
Condition	<i>LM</i>	Site-type	-2.148	1.566	1, 17	F = 1.883	0.188
		Length	-1.194	0.576	1, 17	F = 4.301	0.054
		Site-type*Length	0.910	0.622	1, 17	F = 2.138	0.162

Table 2.5 Summary of results from Linear Models (LM) and Linear Mixed Models (LMM) assessing differences between wastewater-affected and reference sites in trophic ecology (i.e., $\delta^{15}\text{N}$, $\delta^{13}\text{C}$), energy storage (i.e., C:N, condition), and growth of Burbot. The random effect of lake was tested for each covariate. If variance estimates approached zero, and singularity was detected, the model was deemed to be overparameterized. In such instances, relationships were analyzed using a linear model (LM) without a random effect. Parameter estimates (β), standard errors (SE), degrees freedom (df), the F-value (F), and significance (p) are shown. Significant differences ($\alpha = 0.05$) are underlined and bold.

Burbot Model			Results				
Covariate	Model Type	Fixed Effect	β	SE	df	F	p
$\delta^{15}\text{N}_{\text{adj}}$	LM	Site-type	-0.402	0.882	1, 24	0.207	0.653
		Length	0.005	0.002	1, 24	5.857	<u>0.023</u>
		Site-type*Length	-0.003	0.003	1, 24	0.913	0.349
$\delta^{13}\text{C}$	LM	Site-type	0.574	1.211	1, 24	0.204	0.655
		Length	-0.006	0.003	1, 24	5.000	<u>0.035</u>
		Site-type*Length	0.001	0.005	1, 24	0.030	0.865
Growth _{abs}	LM	Site-type	45.853	19.215	1, 25	5.694	<u>0.025</u>
		Length	0.001	0.046	1, 25	<0.001	0.983
		Site-type*Length	-0.063	0.071	1, 25	0.793	0.382
C:N	LM	Site-type	0.246	0.066	1, 24	13.768	<u>0.001</u>
		Length	<-0.001	<0.001	1, 24	1.331	0.260
		Site-type*Length	<-0.001	<0.001	1, 24	6.441	<u>0.018</u>
Condition	LM	Site-type	-0.581	0.461	1, 14	1.590	0.228
		Length	-0.002	0.001	1, 14	1.702	0.213
		Site-type*Length	0.002	0.002	1, 14	1.252	0.282

Nutrient additions from municipal wastewater are known to affect fish health (Munkittrick, 1992; Vadeboncoeur et al., 2001). In the Baker Lake wastewater wetland, Arctic grayling and Burbot from wastewater-affected sites had higher condition, lipid content, and absolute growth rate compared to fish from reference sites, with significant differences observed in Arctic grayling (LMM, $\chi^2(1) = 4.046$, $p = 0.044$) and Burbot (LM, $F(1, 25) = 19.215$, $p = 0.025$) growth

rate, and Burbot lipid content (LM , $F(1, 24) = 6.411$, $p = 0.018$) (Table 2.4 and Table 2.5). Monitoring fish health offers a cost-effective means to gauge the impact of wastewater effluent on ecosystems and aligns with the goals of the Canadian Environmental Effects Monitoring (EEM) program, which aims to maintain healthy fish populations and diverse aquatic ecosystems (Kilgour et al., 2007). The recently established Northern-specific Wastewater Standards (CSA W203:19) reflect similar goals to the EEM program, while also calling for community consultation, integration of traditional knowledge, and adherence to federal or territorial regulations.

2.3.3 Comparison of metal concentrations in fish muscle tissue between wastewater-affected and reference sites

As discussed above, municipal wastewater can release significant quantities of metals into the environment, and the chemistry of municipal wastewaters can enhance bioavailability of metals through metal speciation, complexation, and precipitation (Heath, 1995; Tessier and Campbell, 1987). Fish, as longer-lived organisms, are particularly well-suited as indicators of wastewater impacts on aquatic ecosystems because they reflect prolonged exposure and integrate effects on lower trophic levels (Parks et al., 1991). In this study, metals that significantly differed between wastewater-affected and reference site-types in abiotic ecosystem components (refer to Table 2.1 and Table 2.3) were analyzed in fish dorsal muscle tissue and compared between site-types. These metals included manganese (Mn), copper (Cu), iron (Fe), and zinc (Zn). Most 2019 fish tissue [Cu] were below the method reporting limit (MRL) for that year ($MRL < 1.744$ ug/g), leading to the exclusion of Cu from further analysis in fish. Methylmercury (MeHg) was added to the analysis in response to community concerns.

Although [MeHg] in water and sediment from wastewater-affected systems were up to eight times higher than in reference systems, Arctic grayling (LMM , $\chi^2(1) = 11.102$, $p < 0.001$) and

Burbot (*LMM*, $\chi^2(1) = 4.299$, $p = 0.038$) from wastewater-affected systems had significantly lower size-adjusted tissue [MeHg] than those from reference systems (Figure 2.8).

The linear mixed-effect models were effective in explaining variation in size-corrected MeHg concentrations in Burbot and Arctic Grayling. Specifically, these models explained 77% of variation in MeHg concentrations for Arctic grayling and 81% of variation in MeHg concentrations for Burbot. For Burbot, the difference between the marginal- R^2 (0.321, fixed effects only) and conditional- R^2 (0.813, fixed and random factors; Table 2.7) values indicates that the random factor – “lake” – explained a substantial amount of variation, and that Burbot [MeHg] differences among lakes should be further explored. Causes of among-lake variation in [MeHg] in both Burbot and Arctic Grayling, as well as residual, unexplained variation, may include specific fish characteristics known to affect bioaccumulation. These include factors such as trophic level and foraging habitat (Kidd et al., 1995a; Power et al., 2002), as well as fish condition, fat content, and growth patterns (Lescord et al., 2018; Moslemi-Aqdam et al., 2022, 2023; Thomas et al., 2020). To further understand the lower [MeHg] observed in wastewater-affected systems compared to reference systems, multiple linear regression analysis was employed. Despite lower [MeHg] in wastewater-affected systems, the absolute value of the difference between site-types (Arctic grayling, 0.096 $\mu\text{g/g}$ wet weight; Burbot 0.166 $\mu\text{g/g}$ wet weight) is relevant to Baker Lake community members, who are concerned about [Hg] in fish downstream of the wastewater wetland; additional analyses to explore causes of variation in fish MeHg were thus warranted.

For Arctic grayling, a multiple linear regression that included $\delta^{15}\text{N}$, $\delta^{13}\text{C}$, and condition factor explained 61% of variation in MeHg concentration observed among individual fish (*MLR*, $F(3, 60) = 34.070$, $p < 0.001$). Growth rate and C:N were included in the initial model but were

removed due to multicollinearity and variance inflation. A similar model was constructed for Burbot, and growth rate, C:N, and condition factor were removed to account for multicollinearity and variance inflation. The final multiple regression model for Burbot included $\delta^{15}\text{N}_{\text{adj}}$ and $\delta^{13}\text{C}$, and explained 43% of variation (MLR , $F(2, 25) = 11.340$, $P < 0.001$). MeHg concentrations in both Burbot and Arctic grayling models were positively related to $\delta^{15}\text{N}_{\text{adj}}$, which is consistent with well-established evidence of bioaccumulation and biomagnification of MeHg in aquatic food chains (see [Driscoll et al., 2013](#); [Kidd et al., 1995a](#)). Also consistent with previous research were negative relationships between MeHg concentrations and $\delta^{13}\text{C}$ for both species, which indicates lower MeHg concentrations in fish that rely more on benthic-derived carbon, an outcome described by many previous studies ([Amundsen et al., 2023](#); [Ch  telat et al., 2011](#); [Karimi et al., 2016](#); [Power et al., 2002](#); [Thomas et al., 2016](#)). This effect was more pronounced in Arctic grayling, where $\delta^{13}\text{C}$ was estimated to have a larger impact on muscle tissue [MeHg] compared to Burbot ([Table 2.7](#)). In addition, Arctic grayling displayed significant $\delta^{13}\text{C}$ variation between site-types ([Table 2.7](#)), unlike Burbot. Overall, it appears that the dietary or habitat use changes made by Arctic grayling in response to municipal wastewater may influence MeHg levels. Some of the potential covariates of MeHg that significantly differed between wastewater-affected and reference sites for Arctic grayling (i.e., growth rate) and Burbot (i.e., growth rate and C:N) could not be included in MLRs due to multicollinearity and inflation concerns, which become amplified with small sample sizes ([Kroll and Song, 2013](#)). However, the effect of growth rate on mercury bioaccumulation in fish is well-established ([Karimi et al., 2007](#); [Lavoie et al., 2013](#); [Simoneau et al., 2005](#)), suggesting that the effects of growth-mediated MeHg dilution warrant separate, dedicated study and analyses in the Baker Lake system. In a simple linear regression, a significant

negative relationship was found between length-standardized muscle tissue MeHg and absolute growth rate for Burbot (LM , $F(1, 27) = 12.820$, $p = 0.001$). The relationship between MeHg and absolute growth rate was similarly negative for Arctic grayling, but was not significant (LM , $F(1, 63) = 3.16$, $p = 0.080$). Higher growth rates were observed at wastewater-affected systems for both species (Table S2.4), suggesting the likelihood of somatic growth dilution, which is when increased body mass outpaces metal accumulation and results in lower mercury concentration in tissues (Karimi et al., 2007).

Table 2.6 Results of Linear Models (LM) and Linear Mixed Models (LMM) comparing metal concentrations in muscle tissue from A) Arctic grayling and B) Burbot between wastewater-affected and reference site-types from Baker Lake, Canada, Nunavut. The adjusted R^2 ($R^2_{(adj)}$) describes the proportion of variance explained once the number of predictors is accounted for. The marginal R^2 ($R^2_{(m)}$) describes the proportion of variance explained by fixed factors and the conditional R^2 ($R^2_{(c)}$) describes the proportion of variance explained by both fixed and random factors. For LM, the test statistic is the F-value (F), and for LMM, it is the chi-squared (χ^2) value. Significance level (α) is set at 0.05.

A) Arctic grayling							
Metal	Model Type	$R^2_{(m)/(adj)}$	$R^2_{(c)}$	df	Test statistic	p	Random Effect (variance $\pm$ SD)
$\text{Log}_{10} [\text{Mn}]$	<i>LMM</i>	0.137	0.615	1	$\chi^2 = 1.554$	0.213	0.017 ± 0.132 , (χ^2 , $p=0.001$)
$\text{Log}_{10} [\text{Fe}]$	<i>LM</i>	0.025	N/A	1, 58	$F = 2.058$	0.119	N/A
$\text{Log}_{10} [\text{Zn}]$	<i>LM</i>	-0.009	N/A	1, 58	$F = 0.445$	0.507	N/A
$\text{Log}_{10} [\text{MeHg}]$	<i>LMM</i>	0.513	0.770	1	$\chi^2 = 11.102$	<0.001	0.020 ± 0.141 (χ^2 , $p<0.001$)
B) Burbot							
Metal	Model Type	$R^2_{(m)/(adj)}$	$R^2_{(c)}$	df	Test statistic	p	Random Effect (variance $\pm$ SD)
$\text{Log}_{10} [\text{Mn}]$	<i>LM</i>	0.023	N/A	1, 25	$F = 1.610$	0.216	N/A
$\text{Log}_{10} [\text{Fe}]$	<i>LM</i>	-0.031	N/A	1, 25	$F = 0.176$	0.678	N/A
$\text{Log}_{10} [\text{Zn}]$	<i>LM</i>	0.101	N/A	1, 25	$F = 3.897$	0.060	N/A
$\text{Log}_{10} [\text{MeHg}]$	<i>LMM</i>	0.321	0.813	1	$\chi^2 = 4.299$	0.038	0.056 ± 0.237 (χ^2 , $p<0.001$)

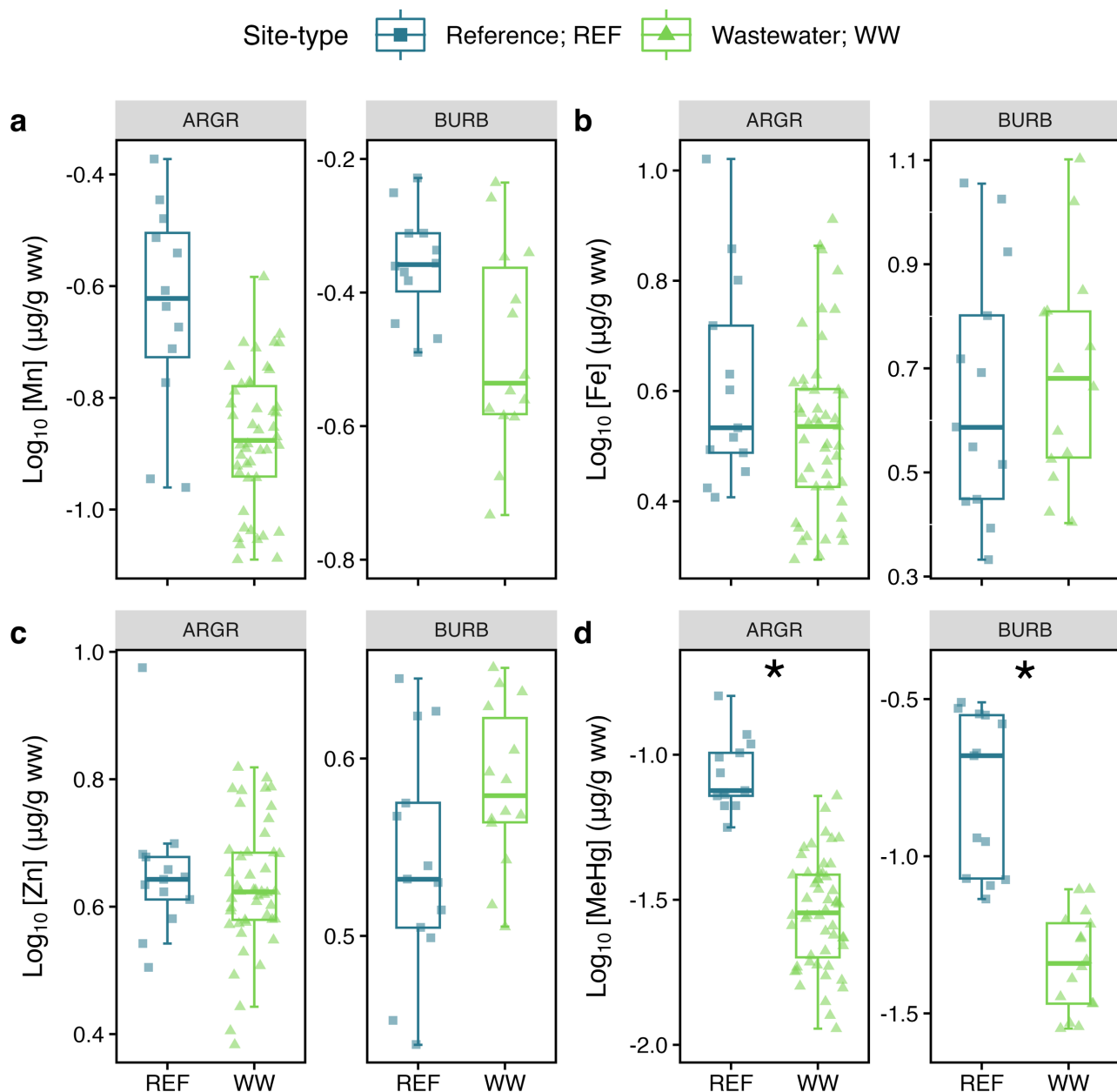


Figure 2.8 Box and whisker plots of size-corrected metal concentrations (µg/g ww; wet weight) ± standard error in muscle tissue for Arctic grayling (ARGR) and Burbot (BURB) from reference (REF, blue squares) and wastewater-affected (WW, green triangles) systems. Linear models (LM) and Linear Mixed Models (LMM) were used to test site-type differences. To account for the effect of body size, residuals from the length – metal concentration relationship were calculated and adjusted to the mean length before modeling. Lake was included as a random factor (wastewater-affected: n=3 lakes; reference: n=2 lakes); when singular bounds were detected, the random factor was removed to avoid over-parameterization. Muscle tissue metal concentrations were significantly lower in Arctic grayling ($LMM, \chi^2(1) = 11.102, p < 0.001$) and Burbot ($LMM, \chi^2(1) = 4.299, p = 0.038$) from the wastewater-affected systems compared to reference systems. Significant difference are signified by an asterisk (*) with alpha set at 0.05.

The limited sample size of this study hinders a thorough assessment of the interactions between lake-specific physicochemistry, fish life-history traits, and MeHg concentrations in fish muscle tissue. The work of [Moslemi-Aqdam et al. \(2022\)](#) provides insights into the complex mechanistic interactions that may influence MeHg levels in the Baker Lake lagoon-tundra wastewater wetland. Their findings indicate that mercury concentrations in top predator fish in a series of subarctic systems are primarily influenced by fish growth rate and MeHg levels in benthic prey, which are, in turn, affected by dissolved organic carbon, water MeHg concentration, and sediment THg levels. [Moslemi-Aqdam et al. \(2022\)](#), among others such as [Karimi et al. \(2007\)](#) and [Wang and Wang \(2012\)](#), suggest that rapid fish growth may mitigate the effects of increased water MeHg in wastewater-impacted areas, leading to lower MeHg concentrations in fish muscle.

While MeHg is toxic and bioaccumulates in the food web, manganese (Mn), iron (Fe), and zinc (Zn) are essential micronutrients. These metals can become harmful when present in excess ([Heath, 1995](#); [Wood et al., 2012](#)) but are metabolically regulated. There were no significant differences in muscle tissue concentrations of Mn, Fe, or Zn between site-types in either Arctic Grayling or Burbot ([Table 2.6](#)). When environmental concentrations of trace metals are elevated, homeostasis can be influenced by competition among metals for absorption or modification of transport proteins ([Chandrapalan and Kwong, 2021](#)). Since no significant differences were noted between wastewater-affected and reference site fish, it may be inferred that increased concentrations of Mn, Fe, and Zn in the wastewater wetland in Baker Lake did not disrupt homeostasis of these micronutrients in either Burbot or Arctic grayling ([Wood et al., 2012](#)).

Table 2.7 Results of a multiple linear regression (MLR) analysis examining the influence of fish life history and trophic ecology variables on methylmercury concentrations ([MeHg]) in a) Arctic grayling and b) Burbot muscle tissue from wastewater-affected (n=3) and reference (n=2) sites. Variation in [MeHg] attributed to fork length was accounted for by using residuals from a preliminary regression. Covariates were included based on criteria set by tolerance (Tol.) and variance inflation factors (VIF) to minimize multicollinearity. The table reports the F-values and significance levels for each predictor.

A) Arctic grayling, 61% of variance explained by $\delta^{15}\text{N}_{\text{adj}}$, $\delta^{13}\text{C}$, and condition factor.

Overall Model	R²_{adj}	SE	df	F	p	Collinearity Statistics	
ARGR[MeHg]	0.612	0.162	3, 60	34.070	<0.001		
Coefficient	Estimate	SE	df	F	P	Tol.	VIF
$\delta^{15}\text{N}_{\text{adj}}$	0.125	0.020	1, 60	37.868	<0.001	0.934	1.071
$\delta^{13}\text{C}$	-0.121	0.021	1, 60	33.277	<0.001	0.998	1.001
log ₁₀ Condition	-1.301	0.315	1, 60	17.099	<0.001	0.935	1.069

A) Burbot, 43% of variance explained by $\delta^{15}\text{N}_{\text{adj}}$ and $\delta^{13}\text{C}$

Overall Model	R²_{adj}	SE	df	F	p	Collinearity Statistics	
ARGR[MeHg]	0.434	0.254	2, 25	11.340	<0.001		
Coefficient	Estimate	SE	df	F	P	Tol.	VIF
$\delta^{15}\text{N}_{\text{adj}}$	0.157	0.059	1, 25	7.106	0.013	0.761	1.314
$\delta^{13}\text{C}$	-0.006	0.002	1, 25	4.580	0.042	0.761	0.761

2.4 Conclusion

Findings from this study address important questions raised by the Baker Lake community concerning the effectiveness of the Hamlet's lagoon-tundra wastewater wetland. More broadly, results help address a serious lack of data from passive, wetland wastewater treatment systems in Arctic environments. I found that concentrations of metals decreased in abiotic ecosystem compartments along the wastewater flow path, reaching similar concentrations to reference systems before discharging into the final receiving environment of Baker Lake. This indicates that

while lakes near the wastewater source have elevated concentrations of metals, the wetland effectively attenuates concentrations over a spatial gradient. This research centered on sampling during the ice-off season and did not examine either seasonal or inter-annual variation in metal concentrations. Such an evaluation would enhance knowledge of system efficacy, especially during freshet when it is known that berms in the Baker Lake wastewater wetland lose integrity and water retention times are typically shorter.

Future monitoring and management should consider the accumulation of metals in wastewater-affected site sediment. Any changes to water chemistry, from effluent overload or system modifications, could disrupt the sediment-water interface and result in leaching of metals sequestered in sediments, such as Hg species and Cu. Copper may warrant special attention due to its changing toxicity with water chemistry and its exceedance of guidelines set for the protection of aquatic life in water and sediment. Additionally, the low dissolved oxygen concentrations observed in Lagoon Lake could hinder the movement of fish between upstream and downstream systems. This connectivity is crucial for species such as Arctic grayling, which rely on both stream and lake habitats during specific life stages.

Elevated metal concentrations in sediment and water from wastewater-affected sites did not result in higher metals concentrations in muscle tissue in either Arctic grayling or Burbot. [MeHg] in muscle tissue was lower in fish from wastewater-affected systems, but differences were not significant, and [MeHg] in muscle tissue for each species was related to isotope-inferred trophic position and carbon source for both species, and additionally to fish condition in Arctic grayling. Both [THg] and [MeHg] in fish muscle tissue were well below consumption guidelines across all study systems and for both species.

This research offers valuable insights into the effects of wastewater inputs on Arctic freshwater ecosystems and the efficacy of passive wastewater treatment in the Baker Lake lagoon-tundra wetland. These findings are increasingly critical as human activities in the Arctic intensify, existing treatment wetlands age, and new regulations are implemented. Considering the planned upgrades to the Baker Lake wastewater treatment system and the overarching goal of sustainable wastewater management in the Arctic, future research should concentrate on further quantifying the temporal variation in key parameters and conducting a more comprehensive investigation of the effects of wastewater inputs on lower trophic levels.

Chapter 3

Beyond Soft Tissues: Otolith Microchemistry as a Potential Monitoring Tool in Arctic Freshwater Ecosystems

3.1 Introduction

Wastewater treatment in the Canadian Arctic mainly relies on passive tundra wetland systems, where wastewater is filtered naturally through networks of lakes and streams. These systems are often chosen for their low maintenance costs and minimal infrastructure needs (Balch et al., 2018; Daley et al., 2015; Hayward et al., 2014; Yates et al., 2020, 2012). However, the effectiveness of these passive systems is constrained by the harsh Arctic climate, prolonged use, climate change, and increasing human activity (Johnson, 2008; Lam and Livingston, 2011). Inadequate wastewater treatment can degrade aquatic ecosystems and impact local water usage. The introduction of organic matter and nutrients from municipal wastewater into receiving waterbodies can lead to eutrophication and seasonal oxygen depletion (Chambers et al., 1997). Moreover, wastewater effluents often contain various contaminants, including metals such as lead (Pb), copper (Cu), and zinc (Zn). Effective watershed management in the Arctic depends on a thorough understanding of the impacts of wastewater inputs on ecosystem ecology and physicochemical properties, a challenge compounded by the frequent lack of comprehensive data and baseline measurements in these regions.

Fish, as higher trophic level organisms, can integrate exposure to contaminants across spatial and temporal gradients. This makes select species excellent indicators of overall ecosystem health, reflecting both abiotic and biotic environmental conditions (Parks et al., 1991). Typically, metal concentrations are measured in the dorsal muscle tissue of fish, providing a

snapshot of recent exposure influenced by tissue- and species-specific uptake and elimination rates ([Kraemer et al., 2005](#); [Trudel and Rasmussen, 1997](#)). However, variability in exposure over the lifetime of a fish cannot be determined using muscle ([Norstrom et al., 1976](#); [Trudel and Rasmussen, 2006](#)), or any other organ that is metabolically active (e.g., liver), but may be elucidated using metabolically inert tissues that continuously accrete, such as otoliths ([Campana, 1999](#)).

All teleost fish have three pairs of otoliths, termed sagittae, lapilli, and asterisci, with sagittae being the largest (e.g., [Campana and Neilson, 1985](#)). In this study, the term “otolith” specifically refers to the sagittal otoliths. Otoliths are primarily comprised of calcium carbonate (CaCO_3), although other forms like calcite and vaterite may also be present (see [Campana, 1999](#)). These calciferous crystal structures are located within fluid-filled sacs of the inner ear and are essential for fish hearing and balance. The fluid, endolymph, acts as a growth medium, providing inorganic materials, proteins, and other accessory macromolecules required for otolith formation and growth ([Payan et al., 2004](#)). Otoliths grow continuously from the embryo stage, through daily deposition of alternating aragonite and protein-rich layers ([Campana, 1983](#); [Pannella, 1971](#)). These layers form annuli, which are represented visually as alternating translucent and opaque bands. Counting annuli from the core (i.e., nucleus) outward allows for estimation of fish age, as each annulus indicates seasonal changes in growth. Under reflected light, each year of growth is represented by distinct zones in the otoliths: one wider, opaque zone indicating faster summer growth, and one narrower, translucent zone that signifies slower winter growth. The complex biological and environmental processes that influence otolith biomineralization may provide a unique opportunity for time-resolved environmental monitoring.

The inert properties of otoliths facilitate development of novel applications related to ecological reconstruction in several sub-disciplines. One example is the development of chemical aging techniques, which are based on correlating zinc (Zn) profile minima with translucent growth zones ([Friedrich and Halden, 2010](#); [Halden et al., 2000](#)). The validation and standardization of these chemical aging techniques remain ongoing, primarily due to an incomplete understanding of what causes the seasonal patterns observed in otolith Zn concentrations ([Heimbrand et al., 2020](#); [Reis-Santos et al., 2023](#)). Otolith microchemistry data have also provided valuable insights into topics such as fish migration, anadromy, stock discrimination, fish provenance, and environmental contamination (e.g., [Elsdon et al., 2008](#); [Friedrich and Halden, 2010](#); [Kennedy et al., 2002](#); [Radtke et al., 1996](#); [Swanson et al., 2010](#); [Zimmerman and Reeves, 2000](#)). There is growing interest in using otolith microchemistry to reconstruct past metal exposure in fish, an approach that requires deeper knowledge, especially regarding how metals are incorporated into otoliths.

Element deposition within otoliths is governed by ion availability during biomineralization ([Barnes and Gillanders, 2013](#); [Izzo et al., 2018](#); [Thomas and Swearer, 2019](#)). Essential components for otolith formation are transported into blood plasma via the gut or gills and subsequently cross the endolymphatic membrane into the endolymph, which surrounds the otolith as it grows (see schematic [Figure S3.1](#)). Elements are then assimilated into the otolith aragonite (CaCO_3) structure, via substitution for calcium (Ca), integration into organic matrix proteins, or filling interstitial space ([Campana, 1999](#)). Elements of similar size to Ca, specifically strontium, lead, and barium, have a propensity to substitute for Ca in otoliths ([Campana, 1999](#); [Radtke and Shafer, 1992](#)). Elements with atomic radii smaller than Ca, such as transition metals like zinc and copper,

typically bind to the otolith matrix proteins. Elements that reside in interstitial spaces, such as sodium and potassium, do not bind to the otolith and are more likely to leach out ([Proctor and Thresher, 1998](#)).

Incorporation of trace elements into otoliths is influenced by many variables. Somatic growth rate, fish age and size, and gonad development are internal factors that impact integration of trace elements into otoliths ([Campana, 1999](#); [Kalish, 1989](#); [Sturrock et al., 2015](#)). For example, [Sturrock et al. \(2015\)](#) observed that reductions in Zn:Ca in otoliths from female fish were inversely correlated to gonadosomatic index (GSI), a measure of gonad weight to total body weight. This indicates that during ovarian maturation, less Zn is integrated into the otolith, possibly due to the role of Zn in vitellogenin production, a vital protein for yolk formation ([Sturrock et al., 2015](#)).

Externally, environmental conditions such as temperature, pH, salinity, dissolved oxygen concentration, and ambient metal concentrations also affect incorporation of trace metals into otoliths (e.g., [Campana, 1999](#); [Elsdon et al., 2008](#); [Elsdon and Gillanders, 2002](#); [Radtke and Shafer, 1992](#)). For example, Sr concentrations are often higher in brackish and saline waters than freshwaters, which has facilitated use of otolith strontium-to-calcium ratios in reconstructions of anadromous fish migrations and discrimination of fresh, brackish, and marine habitat use (e.g., [Zimmerman, 2005](#)). Additionally, the use of multi-element otolith signatures, which can reflect underlying geology and groundwater inputs, can be used to help delineate fish stocks ([Campana, 2005](#); [Thresher, 1999](#)). Although otolith microchemistry serves as a powerful tool for elucidating the complex life histories of fish, the multitude of factors influencing element incorporation introduces significant complexity to interpretation and application.

Recent advancements have highlighted the need for more field-based studies of both intrinsic and extrinsic factors that affect otolith microchemistry ([Grammer et al., 2017](#); [Hüssy et al., 2021b](#); [Reis-Santos et al., 2023](#); [Thomas and Swearer, 2019](#)). After three decades of study, knowledge gaps of otolith microchemistry as an ecological tool have been narrowed; however, the utility of otolith microchemistry in reconstructing past environmental conditions and other applications is only partially understood. Researchers are urged to consider study-level covariates and biomineralization processes ([Hüssy et al., 2021b](#); [Izzo et al., 2018](#); [Sturrock et al., 2014](#)), which are crucial for effectively reconstructing environmental histories.

The subarctic Barrenlands of the Kivalliq Region, characterized by dense networks of low-productivity waterbodies, present an excellent opportunity for research on otolith microchemistry. These lakes, classified as oligotrophic or ultra-oligotrophic, are sensitive to environmental changes and serve as early indicators of ecological disturbances (e.g., [Goedkoop et al., 2022](#); [Huser et al., 2022](#)). Specifically, the Baker Lake lagoon-tundra wastewater wetland offers a unique setting to study otolith microchemistry as an inland Arctic freshwater system that has been subject to eutrophication and metal contamination from municipal wastewater.

In [Chapter 2](#), metal concentrations and indicators of primary productivity were found to differ between lakes affected by wastewater and unimpacted lakes. This prompted a deeper investigation into otolith microchemistry across a subset of lakes, which represent upstream, wastewater-affected, and reference site-types. To ensure the reliability of comparisons, lakes were selected based on having similar size-class distribution of captured fish. The lakes included in this chapter included: 1) Upper Lagoon Lake, located immediately upstream of the first wastewater lake in the lagoon-tundra treatment system; 2) Airplane Lake, the most downstream

lake in the lagoon-tundra wastewater wetland before discharge into the greater Baker Lake basin; and 3) R1, a reference lake not affected by wastewater.

The core hypothesis of this chapter posits that metal concentrations in otoliths will vary among lakes and will correlate with metal concentrations in soft fish tissues, such as muscle and liver. This hypothesis frames subsequent analysis where I investigated the relationship between metal concentrations in otoliths and soft tissues, considering both internal and external factors that may influence metal incorporation into otoliths. I measured and compared metal concentrations in the first-year growth and marginal edges of otoliths among the three studied lakes, related concentrations in otoliths to those found in soft tissues, and related concentrations to possible covariates. I used results to assess the capacity of otolith microchemistry to chronologically reconstruct contaminant exposure. Through this research, I hope to further knowledge regarding the application of otolith microchemistry as a tool for reconstructing metal exposure, with applications to systems, such as those in the Arctic, where baseline data are limited.

3.2 Methods

3.2.1 Study Site and Sample Collection

During summers of 2019 and 2021, Arctic grayling (*Thymallus arcticus*) were captured in three lakes near the hamlet of Baker Lake (64.32°N, 96.02°W) in Nunavut's Kivalliq region, Canada (Figure 3.1). These lakes, all located within 3 km of Baker Lake, included Upper Lagoon Lake, situated upstream and potentially influenced by wastewater, Airplane Lake, located downstream of influent, and Lake R1 (Reference 1). Lake R1 was selected for its geological similarity to Airplane Lake and its accessibility. Between 14 and 29 fish were captured in each lake using

various methods, including 2-hour small-mesh (20 mm) gill net sets, 12-hour fyke net sets, and angling from the shore in accordance with University of Waterloo Animal Care guidelines (AUP #41176). Sampling occurred during the open-water season and targeted various habitats within each lake (i.e., methods were not standardized across lakes). The fish were measured for length (in millimeters) and weight (in grams) before being sent frozen to the University of Waterloo for further analysis.

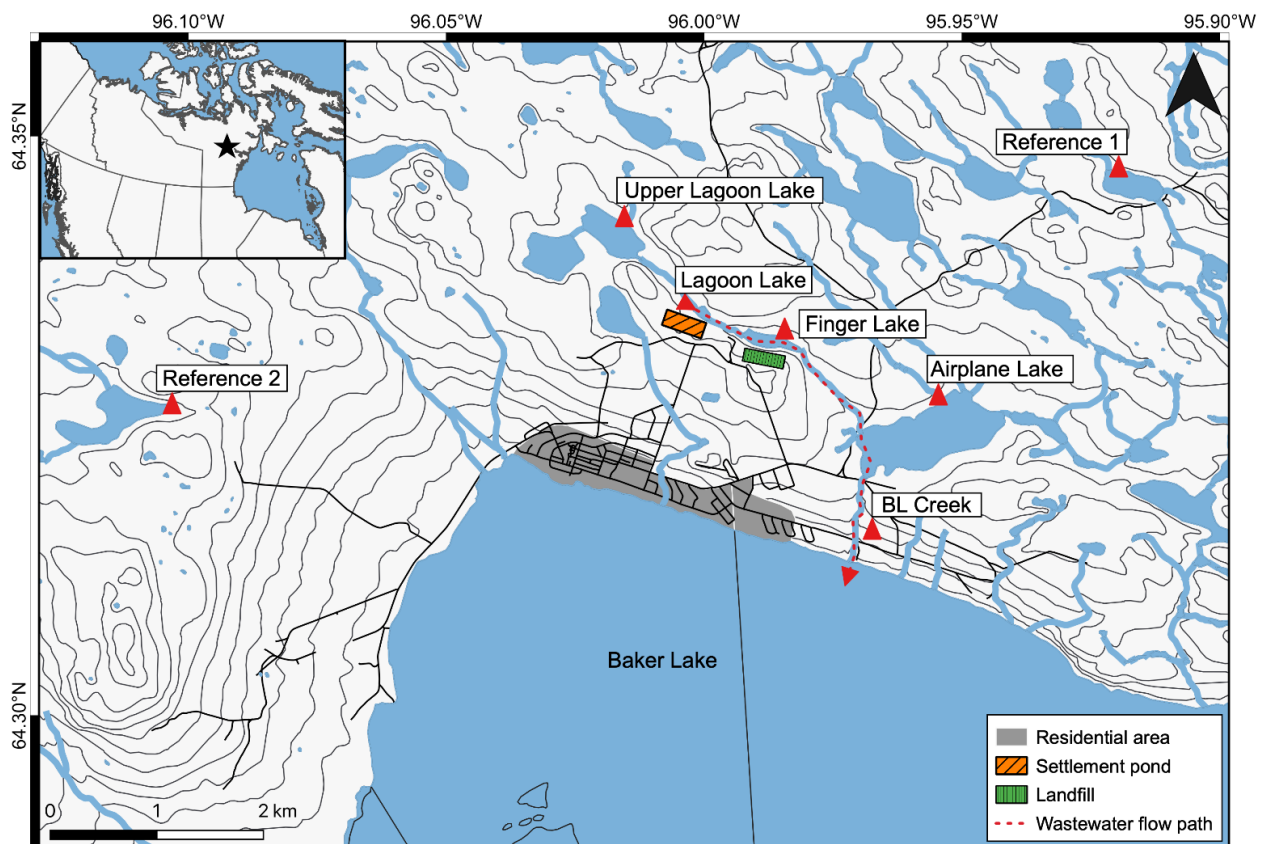


Figure 3.1 Study lakes in Baker Lake, Nunavut, Canada. A subset of three lakes were sampled for otolith microchemistry analysis, including Upper Lagoon Lake, Airplane Lake, and Reference 1 (R1).

3.2.2 Sample processing

At the University of Waterloo Fish Lab, large-bodied fish dorsal muscle tissue with skin removed was obtained from the left and right side of each fish. An incision was made on the ventral side of the fish starting at the anus and cutting to the base of the gills. Once the internal organs were exposed, sex and maturity of each fish were determined. Gonads and livers were removed and weighed. Sagittal otoliths were removed by cutting through the skull and exposing the brain cavity. The membranous sac surrounding the otolith was removed before drying and subsequent storage in an envelope to ensure no moisture would degrade the sample.

Dorsal muscle tissue was freeze-dried using a LabConco FreeZone drier set at -54°C and 0.014 mBar until the sample was void of moisture. Dry samples were ground by motor and pestle until homogenized. The high fat content of fish livers precludes complete moisture removal by freeze-drying; thus, livers were frozen with liquid nitrogen (N₂) and pulverized in a Cryo-Cup Grinder (BioSpec Products, Bartlesville, OK, USA) until homogenized.

Homogenized muscle and liver were analyzed at the Biotron Center for Experimental Climate Change Research (University of Western Ontario, London, ON, CA), an ISO 17025 accredited laboratory. Total metal concentrations (i.e., Mn, Fe, Cu, Zn, Pb) were determined following EPA Methods 3052 and 200.8 ([U.S. EPA, 1996b, 1994](#)). Metals were selected for analysis based on a literature review of trace elements found in municipal wastewater that have previously been recorded in otoliths (see [Table S2.1](#)).

3.2.3 Otolith Preparation

To prepare otoliths for microchemistry analysis, the right sagittal otolith of each fish was embedded, sulcus side up, in epoxy (Cold Cure™ 2:1 ratio). If the right otolith was not collected,

the left otolith was used. Epoxy was mixed and sonicated to ensure that it was free of air bubbles that could interfere with laser ablation. Embedded otoliths cured for 72 hours at room temperature until the epoxy had fully hardened. Otoliths were then sectioned (~2.5 mm) transversely with a low-speed IsoMet (Buehler Ltd, Illinois) diamond blade saw, such that the nucleus (i.e., origin of growth) was included. Sections were polished on a series of abrasive polishing papers with decreasing grit sizes (i.e., 100, 30, and 3 μm) until smooth, removing any surface irregularities that could affect analysis.

Five to seven individual otolith cross sections were mounted flat in a 25 mm Lucite microprobe mount. The Lucite disk was backfilled with epoxy and left to cure for 72 hours. Once cured, disks were wet-polished using deionized water and 100, 30, and 3 μm polishing paper, followed by a Buehler MetaServ250 Grinder-Polisher with Buehler MasterPolish (0.05 μm). Otoliths were frequently assessed under a microscope during grinding and polishing to ensure nucleus integrity. Polished disks were ultrasonically cleaned with deionized water for 15-minutes and individual cross sections were imaged using a Leica M80 dissecting microscope with a Leica IC80 camera attachment under reflected light.

3.2.5 Age determination

Estimations of age were conducted with a dissecting microscope and camera attachment by counting pairs of opaque and translucent bands (i.e., annuli) on the exposed otolith surface. The outer boundary of each translucent zone was considered the formation point for the next annulus, representing one year of incremental growth. Otolith images were captured and processed using LAS X microscope software (Leica Microsystems, Switzerland).

The accuracy of age determination was evaluated using a scoring system and multiple assessments of age. Each age estimate was assigned a confidence level (by the reader) on a scale of 1 to 10, with higher scores indicating greater confidence. An age estimate was deemed valid if both the age and the confidence rankings were consistent across three readings of each otolith. In cases where the age estimates did not match or the confidence scores were low (i.e., <5), a second interpreter conducted independent assessments, and the results were compared between interpreters until consensus. This method is described by [Casselman \(1983\)](#).

3.2.4 LA-ICPMS of Otoliths

Laser ablation inductively coupled plasma mass spectrometry (LA-ICP-MS) was conducted at the Department of Earth Sciences, University of Manitoba, Winnipeg, with an Element 2 ICP-MS (Thermo Finnigan, High Resolution) coupled to a New Wave laser (UP-213, Merchantek, Fremont, California). Typical operating conditions are summarized in [Table S3. 1](#). Calcium (Ca^{48}) was used as an internal standard (56% CaO, g/mol), and NIST-SRM-610 (glass) standard reference material (National Institute of Standards and Testing) was used as an external standard. NIST-SRM-610 was spot ablated (5 Hz, 30 μm , 70% power) before and after ablations were performed on each disk (5-7 samples). A pre-ablation transect was run before elemental assay to remove any surface contaminants ([Campana et al., 1994](#); [Thresher, 1999](#)). Subsequently, continuous transects were ablated from each otolith nucleus to the outer dorsal edge. Otolith material was vaporized by the laser and transported by argon gas to the ICP-MS for analysis of elemental concentrations ([Denoyer et al., 1991](#)). Trace element concentrations were processed in the data reduction software GLITTER ([van Achterbergh et al., 1999](#)) and Iolite ([Paton et al., 2011](#)); intensities were converted to parts per million (ppm), and reported relative to Ca^{48} .

Time series data of metal concentrations in otoliths were smoothed using a 10-point moving average and plotted against distance travelled by the laser (μm) (calculated as speed of the laser in $\mu\text{m}/\text{second} \times \text{ablation time in seconds}$, from otolith core to distal end). Mean metal concentrations were then determined for two increments of interest for otolith growth: 1) year-one (i.e., first full year of growth after hatch) and 2) marginal edge (i.e., last full year of growth before capture).

3.2.5 Statistical Analysis

Univariate and multivariate techniques were used to test hypotheses concerning individual elements and multi-element signatures. Univariate normality was examined as part of preliminary data exploration. A Shapiro-Wilk test was used to examine the assumption of normality for all study parameters by computing sum-of-squares differences between observed and expected values. Data were $\log_{10}$ -transformed as required. Normality of residuals was assessed using visual inspection of residual distributions and Q-Q plots. Alpha was generally set to 0.05, with exceptions explicitly stated. All statistical analyses were conducted in RStudio version 2023.12.1+402 using core packages from R version 4.3.3 (2024-02-29), unless otherwise indicated.

Pearson correlation analyses were conducted to examine the strength and direction of linear relationships between otolith metal concentrations (i.e., Mn, Fe, Cu, Zn, Pb) and fish life history characteristics (i.e., age, length, weight, condition, GSI, LSI, growth rate, C:N, $\delta^{13}\text{C}$, $\delta^{15}\text{N}$). The analyses were conducted twice, once on year-one data from otoliths, and once on marginal edge data from otoliths. Pearson Correlation matrices were calculated using the `'recorr()'` function in the R package *Hmisc* (Harrell Jr, 2019). Additionally, correlations between

concentrations of metals in otolith marginal edges and soft tissues (i.e., muscle and liver) were examined. Correlations between concentrations of metals in year-one otolith increments and soft tissues were not conducted because year-one metal concentrations are not reflective of recent exposure.

Before examining differences in otolith microchemistry among the three study lakes, the potential influence of fish size on element concentrations was investigated, as suggested by [Campana \(2005\)](#). Initial Pearson correlation tests between fish size and metal concentrations in the first-year otolith increments indicated no significant relationships ($p > 0.05$); therefore, no adjustments for fish size were made for these data. However, significant correlations between fish length and concentrations of Mn, Zn, and Fe were observed at the marginal edges in at least one lake— Upper Lagoon Lake ([Table S3.3](#)), Airplane Lake ([Table S3.5](#)), and Lake R1 ([Table S3.7](#)). To address these correlations, simple linear regressions of metal against fork length were computed independently for each metal and lake. Residuals from these regressions were extracted and added to the mean metal concentration, effectively correcting for the effect of size ([Campana, 2005](#); [Gotelli and Ellison, 2013](#)). These length-adjusted marginal edge metal concentrations were utilized in subsequent analyses.

A permutational multivariate analysis of variance (PerMANOVA) was used to test for differences in otolith microchemistries among lakes (i.e., group centroids). Assumptions of multivariate normality and homogeneity of variance were considered, although PerMANOVA is robust to violations in these assumptions ([Anderson, 2001](#)). Multivariate normality was assessed by analyzing Q-Q plots and comparing skewness (i.e., asymmetry) and kurtosis (i.e., the weight of tails) scores to normal multivariate distributions. Homogeneity of variance was evaluated using

Bartlett's Test, which tests the null hypothesis of equal group variance under a chi-square distribution (Bartlett, 1937). A dissimilarity matrix was generated using the *vegan* package in R (Oksanen et al., 2019) by calculating differences in multi-element otolith signatures between each pair of fish based on Euclidean distance (Gower, 1966). This metric is particularly suitable for analyzing continuous data, such as otolith metal concentrations, as it quantifies the absolute magnitude of distance and treats all dimensions (i.e., each metal element in the otolith signature) equally (Anderson, 2001). The test statistic was then calculated as the ratio of the sum of squared Euclidean distances among groups divided by the sum of squared distances within groups (i.e., pseudo-F statistic and *p*-value) (Anderson, 2001). PerMANOVA was performed using the '*adonis2()*' function by randomly rearranging (i.e., permutating) the dissimilarity matrix 1000 times and testing the null hypothesis of no differences in metal concentrations among lakes in year-one and marginal edge growth increment in otoliths.

Principal component analysis (PCA) served as the ordination method. Dissimilarity matrices were derived from log₁₀-transformed data of year-one and length-adjusted marginal edge values using Euclidean distances in the R package *factoextra* (Kassambara, 2016). Variable contributions (in percentage) to principal components were assessed using squared cosine (cos²) values, which measure representational quality based on angular relationships in right triangles (e.g., Abdi and Williams, 2010).

To explore differences in individual mean metal concentrations among lakes, analyses of variance (ANOVAs) were performed on log₁₀-transformed data of year-one and length-adjusted marginal edge values. Pairwise comparisons of significant ANOVA results were evaluated with

Tukey's Honestly Significant Difference (HSD) post-hoc test, with p -value adjustments for multiple comparisons.

3.3 Results and Discussion

The utility of otoliths has expanded beyond traditional age determination, and otoliths can now serve, in some circumstances, as tools for answering biological and ecological questions related to movements and habitat use of fish (see [Izzo et al., 2018](#); [Reis-Santos et al., 2023](#)). Otolith microchemistry can provide a unique temporal perspective of exposure history that surpasses the capabilities of traditional soft tissue-based analyses in environmental assessments. For otoliths to accurately reflect environmental conditions, it is essential to consider both external factors, such as ambient water metal concentrations and temperature, as well as internal factors, including diet and somatic growth, that influence trace element concentrations in otoliths. In this research, complete time-resolved profiles of metals concentrations were generated, with analyses focusing on two crucial growth periods: the initial year of growth, and the most recent annual growth at the marginal edge. Analyses focused on five metals, four of which are micronutrients (Zn, Mn, Cu, Fe) that are metabolically regulated and essential but toxic at high concentrations, and Pb, which is a toxicant only. Concentrations of each metal were compared among study systems and related to a series of intrinsic covariates that represent trophic ecology (i.e., carbon and nitrogen stable isotope ratios), energy storage (e.g., gonadosomatic index), and life history (e.g., length and age).

Because concentrations of individual elements can reflect environmental or physiological processes affecting uptake in ways that are specific to each element, it is often beneficial to first analyze and interpret multi-element otolith signatures, particularly when intrinsic regulatory

processes may mask environmental signals (Reis-Santos et al., 2018; Sturrock et al., 2015). In this study, multivariate analyses revealed that otoliths from a lake receiving primary treated municipal wastewater (i.e., Airplane Lake) had distinct multi-element (i.e., Mn, Fe, Cu, Zn, Pb) otolith signatures compared to an upstream (i.e., Upper Lagoon Lake) and reference lake (i.e., Lake R1) (Figure 3.2) There was a greater difference in otolith microchemistry among lakes when comparing marginal edge increments (*PerMANOVA*, $F(2, 50)=4.1822$, $p=0.002$) rather than year-one increments (*PerMANOVA*, $F(2, 48)=4.038$, $p=0.012$), but both were significant.

Pairwise comparisons of beta dispersion distances revealed that marginal edge microchemistry was significantly different between Airplane Lake and Upper Lagoon Lake

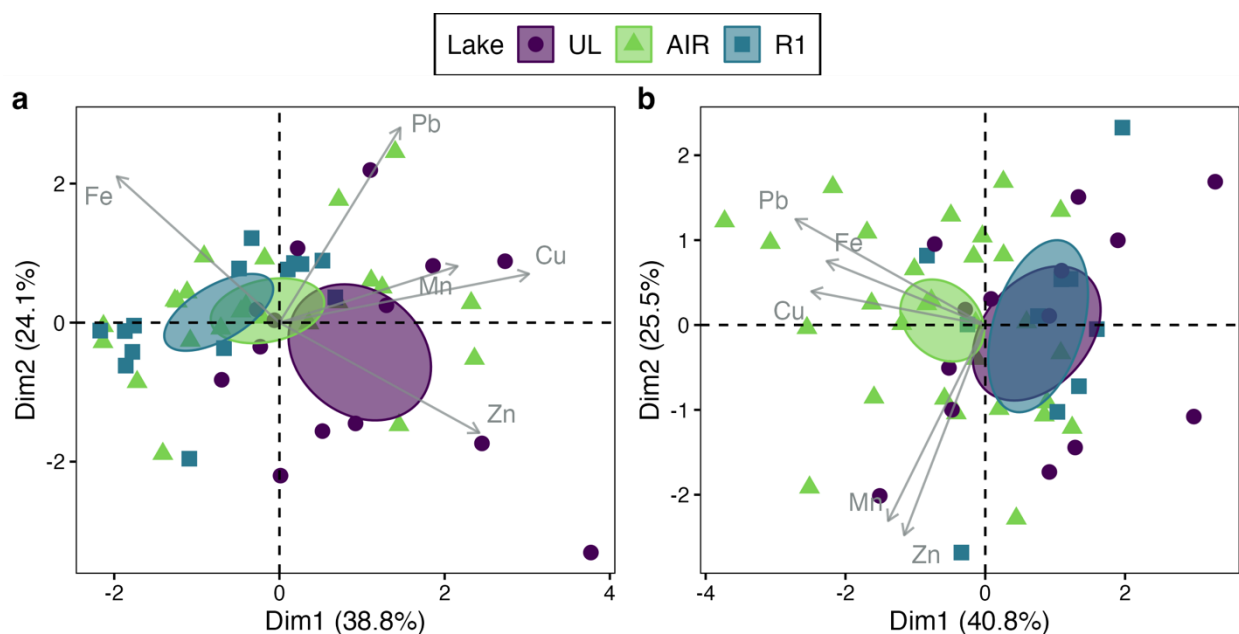


Figure 3.2 Principal component analysis (PCA) biplots for a) year-one otolith signatures and b) marginal edge otolith signatures. Each point on the plot signifies an individual fish metal signature based on measurements of five metals (Fe, Pb, Mn, Cu, and Zn) quantified using ICP-MS. Study lakes include Upper Lagoon Lake (circles), upstream of wastewater input; Airplane Lake (triangles), receives wastewater; and, the reference, Lake R1 (squares). Elemental signatures of both year-one and marginal edge differed significantly among the fish from the three study lakes, although marginal edge otolith composition (*PerMANOVA*, $F(2, 50)=4.1822$, $p=0.002$) was more distinct than year-one (*PerMANOVA*, $F(2, 48)=4.038$, $p=0.012$). Lead (Pb) contributed the most to among-lake variance in metal signatures for both year-one and marginal edge otolith regions.

(*Tukey's*, $p=0.027$), and between Airplane Lake and Lake R1 (*Tukey's*, $p=0.015$). There was no significant difference in marginal edge microchemistry between the two systems not affected by wastewater - Upper Lagoon Lake and Lake R1 (*Tukey's*, $p>0.05$). A post-hoc analysis of year-one otolith microchemistry showed a small but significant difference between Airplane Lake and Upper Lagoon Lake (*Tukey's*, $p=0.048$). There was no significant difference in year-one otolith microchemistry between Lake R1 and either Airplane Lake or Upper Lagoon Lake (*Tukey's*, $p>0.05$). This lack of distinction in year-one increments, compared to marginal edge increments, could reflect variation in relative influence of internal versus external influences on otolith chemistry at different life stages, potentially related to the ratio of aragonite to protein in annuli ([Thorrold and Swearer, 2009](#)).

Results show that for both year-one and marginal edge multi-element signatures, [Pb] had a strong influence on microchemical differences among lakes, as evidenced by its contributions to overall PCA dissimilarity ([Table S3.9](#) and [Table S3.10](#)). This may reflect higher [Pb] levels in sediments of Airplane Lake relative to the other lakes (see [Chapter 2](#); [Figure 2.6](#)), although single-element analysis revealed no significant differences in otolith [Pb] among the three lakes studied in this chapter ([Table 3.1](#)). Authors of a study on a river polluted by industrial activities noted site-specific variations in otolith [Pb] ([Ranaldi and Gagnon, 2008b](#)), and found a relationship between [Pb] in sediment and [Pb] in otoliths ([Ranaldi and Gagnon, 2008a](#)). Drawing from this, [Thomas et al. \(2017\)](#) propose that Pb might serve as a marker for environmental contamination in waterbodies, but only if levels are high enough to overcome any internal physiological elimination processes. Concentrations of other elements also have to be considered. Mechanisms of Pb transport and incorporation differ from those of essential

elements; Pb exists as a free ion in the otolith matrix but is bound to proteins in the endolymph (Izzo et al., 2015; Mugiya et al., 1991; Thomas et al., 2017). Significant negative correlations between [Pb] and [Fe] were observed in Upper Lagoon Lake (Pearson, $r=-0.605$, $p=0.022$), and between [Pb] and [Cu] in Airplane Lake (Pearson, $r=-0.464$, $p=0.011$) and Lake R1 (Pearson, $r=-0.803$, $p<0.001$). These inverse relationships suggest that increases in [Pb] may lead to decreases in other metal concentrations and vice versa, potentially through competitive binding within the otolith matrix (Thomas et al., 2017), the endolymph, or both.

Table 3.1 Mean $\pm$ standard error metal concentrations (in parts per million; ppm) in otoliths of Arctic grayling from three lakes near Baker Lake, NU. Metal concentrations are reported for two regions of otolith growth: 1) year-one (i.e., first full year of growth after hatch) and 2) marginal edge (i.e., last full year of growth before capture). Marginal edge values are corrected for body size by adding residuals of the length – [metal] linear regression to mean [metal]. Significant pairwise differences of ($\log_{10}$ -transformed) metal concentrations are depicted with compact letter display (Tukey's HSD on ANOVA, $p<0.05$) and bold. ANOVA results reported in Table S3.8.

Mean [metal]	Upper Lagoon Lake	Airplane Lake	Lake R1
1) Year-one, n	14	29	14
Mn (ppm)	5.041 (0.623)	4.402 (0.463)	5.222 (0.651)
Fe (ppm)	4.989 (0.892) ^A	9.540 (1.056) ^B	7.488 (0.814) ^{AB}
Cu (ppm)	0.821 (0.113) ^A	0.761 (0.119) ^{AB}	0.461 (0.089) ^B
Zn (ppm)	130.313 (11.972) ^A	108.416 (8.142) ^{AB}	74.489 (4.617) ^B
Pb (ppm)	0.022 (0.009)	0.016 (0.004)	0.013 (0.002)
2) [Length-adjusted] Marginal edge, n	14	29	10
Mn (ppm)	2.517 (0.348)	3.133 (0.490)	3.597 (0.591)
Fe (ppm)	6.978 (1.753) ^A	15.553 (2.121) ^B	6.657 (0.840) ^A
Cu (ppm)	0.332 (0.064)	0.911 (0.230)	0.405 (0.159)
Zn (ppm)	59.950 (15.970) ^A	57.579 (9.681) ^{AB}	21.922 (4.970) ^B
Pb (ppm)	0.013 (0.002)	0.048 (0.028)	0.040 (0.015)

In a recent study of multi-element otolith signatures, [Thomas et al. \(2020\)](#) used cluster analysis to explore the effect of categorizing elements based on their biomineralization processes on determining the natal origins of fish. They found that categorizing elements as associating more with either salt or protein fractions improves the accuracy of origin determination. Specifically, Mn was identified as protein-dominant in the otolith nucleus but salt-dominant (i.e., CaCO_3) during the larval phase, due to its presence in both the protein-rich core and the salt fractions of annuli. This detailed analysis revealed that elements associated with salt fractions are particularly effective in distinguishing the natal origin of fish. Future research could extend the techniques of [Thomas et al. \(2020\)](#) by investigating an *a priori* selection of elements based on otolith biomineralization processes influenced by both internal and external factors across different life-history stages, aiming to refine methods and elements selected for reconstructing historical contaminant exposure.

Elements under physiological control, such as transition metals (e.g., Cu, Zn, Mn, Fe), are assumed to provide limited information about environmental exposure ([Hüssy et al., 2024](#); [Thomas et al., 2020](#)) but may still be useful in an otolith microchemistry context when studying highly polluted areas ([Thomas and Swearer, 2019](#)). I found that [Fe] in marginal edge increments were significantly higher in Airplane Lake, a wastewater receiving waterbody, compared to Upper Lagoon Lake and Lake R1, which do not receive wastewater (*Tukey's*, $p < 0.05$; [Table 3.1](#)). In year-one increments, [Fe] were significantly higher in Airplane Lake compared to Upper Lagoon, but not R1. These findings align with significantly elevated [Fe] in water of the Baker Lake wastewater wetland (see [Chapter 2](#); [Table 2.1](#)), and indicate that otoliths did record elevated ambient [Fe] in Airplane Lake. Authors of two recent studies found evidence of an iron-binding protein,

Transferrin (also called *Serotransferrin*), in the otolith matrix (Thomas et al., 2019; Weigele et al., 2016). Thomas et al. (2019) also reports extremely high levels of *Transferrin* in the endolymph, and results of both these studies indicate that presence of Fe in otoliths may not be entirely related to the function of iron as an internally regulated protein cofactor. Otoliths may therefore “record” elevated [Fe] in more polluted waters but be less reliable indicators when variability among systems (or over time) are reduced and intrinsic factors dominate incorporation (Izzo et al., 2018). Overall, it appears that use of otolith [Fe] as a marker of municipal wastewater exposure would need to be assessed on a case-by-case basis.

Few studies have explored correlations between metal concentrations in otoliths and metals concentrations in soft tissues – such as liver and muscle - that are commonly used in environmental monitoring and ecotoxicology. It remains unclear whether relationships between metal concentrations in otoliths and soft tissues vary across different environments (Barnes and Gillanders, 2013; Hansson et al., 2020). To address this knowledge gap, Pearson’s correlation analysis was used to compare metals concentrations in otolith marginal edges with corresponding concentrations in the muscle tissue and liver of individual fish (Table S3.2, Table S3.4, Table S3.6). The results revealed no significant relationships, except for a negative correlation between Lake R1 liver and otolith [Zn] (Pearson, $r=-0.583$, $p=0.041$). A lack of correlation between otolith marginal edge and soft tissue concentrations for the other metals tested (refer to Table S3.2, Table S3.4, Table S3.6) may be due to differences in metabolic processes in otoliths compared to other tissues (Hüssy, 2008; Vignon et al., 2023), as well as the complex processes that govern biomineralization and metal uptake into the otolith matrix, which

involves transport of dissolved ions across multiple membranes from surrounding water and food sources ([Campana, 1999](#)).

To explore intrinsic factors influencing otolith composition, fish life-history characteristics (e.g., age, growth, fork length, weight) were correlated with metal concentrations in both marginal edge and year-one otolith increments. Output tables for Pearson correlations are performed on metals concentrations in otolith marginal edges are reported in [Table S3.3](#), [Table S3.5](#), and [Table S3.7](#). No significant relationships were noted between year-one otolith metal concentrations and fish life-history characteristics (*Pearson*, $p>0.05$). In the marginal edge increments, [Zn] was significantly and negatively related to fork length in all three lakes Upper Lagoon Lake (*Pearson*, $r=-0.678$, $p=0.008$), Airplane Lake (*Pearson*, $r=-0.695$, $p<0.001$), and Lake R1 (*Pearson*, $r=-0.563$, $p=0.009$). Additionally, [Zn] in marginal edge increments were significantly and negatively related to gonadosomatic index, a measure of gonad to body weight ratio, in Airplane Lake [Zn] (*Pearson*, $r=-0.649$, $p<0.001$) and to otolith [Mn] in Lake R1 (*Pearson*, $r=-0.747$, $p<0.001$). Marginal edge Mn concentrations were significantly and negatively related to fork length in both Airplane Lake (*Pearson*, $r=-0.656$, $p<0.001$) and Upper Lagoon Lake (*Pearson*, $r=-0.667$, $p=0.009$). Weaker, yet significant, positive relationships were observed between marginal edge [Fe] and fork length in Airplane Lake (*Pearson*, $r=-0.426$, $p=0.021$) and between marginal edge [Fe] and fish age in Lake R1 (*Pearson*, $r=-0.528$, $p=0.016$).

Otolith profiles from all lakes demonstrated annular peaks and troughs of [Mn] and [Zn] ([Figure 3.3](#)). In all study lakes, the concentrations of [Mn] and [Zn] were higher in the first-year growth increments than in the marginal edge increments, as detailed in [Table 3.1](#). These patterns, along with the significant negative correlations between fish length and the gonadosomatic index

with both [Zn] and [Mn] (Table S3.3, Table S3.5, and Table S3.7), align with previously documented ontogenetic trends, wherein concentrations of Zn (Friedrich and Halden, 2010; Papadopoulou et al., 1978; Ranaldi and Gagnon, 2008b) and Mn (Friedrich and Halden, 2010; Limburg et al., 2015) decrease from the nucleus to the outer edge.

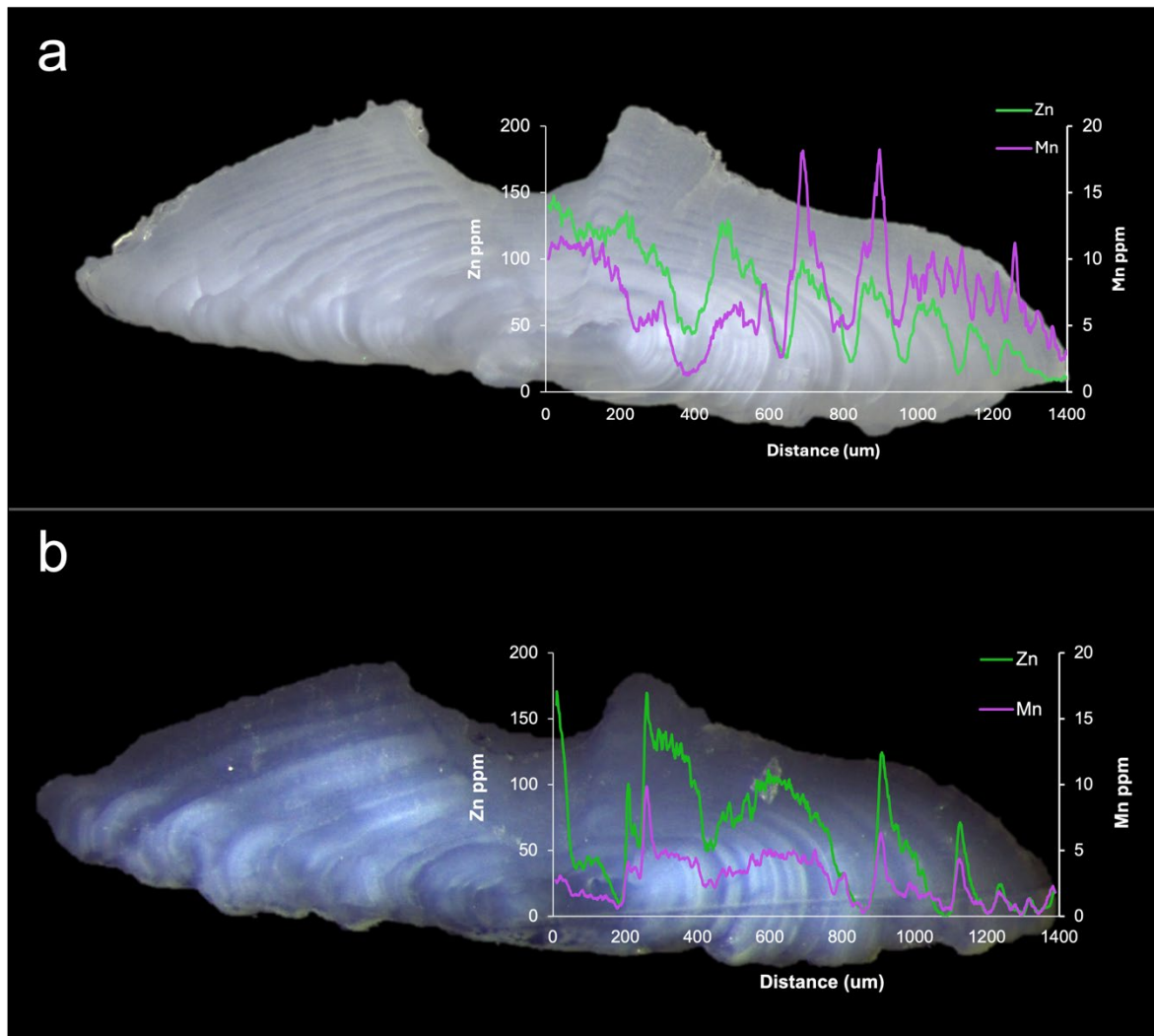


Figure 3.3 Plots of Arctic grayling otolith Zn and Mn concentrations (ppm) smoothed ($n=10$) and plotted over distance (μm). Oscillatory patterns of [Zn] and [Mn] align with annular growth structure in otoliths from both: a) a lake downstream of wastewater input (i.e., Airplane Lake) and b) a reference lake not connected to the wastewater wetland (i.e., Lake R1).

Elevated zinc concentrations in the center of otoliths are thought to reflect increased organic content, specifically proteins, in the otolith nucleus. There is some evidence suggesting that Zn is incorporated into otoliths exclusively within the proteinaceous fraction ([Miller et al., 2006](#)), where it serves as a cofactor for several otolith growth proteins. For example, the enzyme carbonic anhydrase requires Zn in the catalytic conversion of carbon dioxide to bicarbonate, a crucial step in aragonite formation ([Tohse et al., 2006](#)). Other researchers have found, however, that Zn can be found in both the proteinaceous and salt fractions of otoliths ([Izzo et al., 2015](#)). Further work on the biomineralization processes that govern [Zn] in otoliths is required.

Dietary intake is the primary source of Zn to otoliths ([Hüssy et al., 2021b](#); [Ranaldi and Gagnon, 2008b](#)), and seasonal changes in diet affect both otolith accretion rate and Zn availability. During active summer feeding periods, otoliths tend to contain higher concentrations of Zn whereas lower concentrations are observed during the limited winter-feeding period ([Halden et al., 2000](#)). The relationship between otolith [Zn] and seasonal variability in growth makes Zn a useful chemical indicator of fish age ([Halden et al., 2000](#); [Ranaldi and Gagnon, 2008b](#)). However, the effectiveness of Zn as tracer of exposure to ambient concentrations is still unclear ([Thomas et al., 2017](#)). Despite the fact that [Zn] were slightly elevated in Airplane Lake sediment relative to reference systems (see Chapter 2), oscillatory patterns in [Zn] continued to be observed in Airplane Lake ([Figure 3.2](#)). It thus appears that seasonal oscillations in Zn are observed in otoliths even when ambient concentrations are elevated due to anthropogenic disturbance, at least to the extent observed in this study.

Similar to Zn, the mechanisms underlying Mn incorporation into otoliths are not entirely understood, although it is proposed that Mn functions as a protein cofactor during the nucleation

phase of otolith formation (e.g., [Limburg et al., 2015](#); [Ruttenberg et al., 2005](#)). Mn has been detected in both the salt and protein fractions of otoliths ([Izzo et al., 2015](#)), with [Miller et al. \(2006\)](#) identifying it exclusively in the salt fraction. [Thomas et al. \(2017\)](#) showed that Mn exists as a free ion in the endolymph and becomes highly enriched in the otolith, where it likely substitutes for Ca in the aragonite lattice due to a shared oxidation state (both 2+) and similar ionic radius ([Campana, 1999](#)). It is possible that Mn binds to proteinaceous fractions during larval development and to salt components thereafter ([Thomas et al., 2017](#)) and may therefore be a more effective tracer of elevated ambient concentrations in regions of the otolith that represent the adult life-stage ([Thomas et al., 2020](#)), but more research is necessary.

Oscillatory patterns in otolith [Mn] have been observed by several authors (e.g., [Hüssy et al., 2021a](#); [Limburg et al., 2015](#)). [Limburg et al. \(2015\)](#) proposed that the oscillatory patterns of Mn observed in some otoliths ([Figure 3.3](#)) may be due to the substitution of Mn for Ca in differing redox conditions. Under anoxic conditions, Mn^{2+} is highly soluble and more effectively competes with Ca^{2+} ; this could allow variability in dissolved oxygen concentrations to be captured in the otolith record. Here, both year-one and marginal edge [Mn] were not significantly different among lakes (ANOVA, $p>0.05$). Despite higher [Mn] in the water of lakes affected by wastewater, such as Airplane Lake (see [Chapter 2](#); [Table 2.1](#)), and results of previous studies (e.g., [Dorval et al., 2007](#)) that indicate that [Mn] in otoliths reflect ambient [Mn], my findings indicate that variables other than ambient concentrations influence [Mn], perhaps including dissolved oxygen – but more research is necessary.

My research indicates that there is potential to understand historical exposure to municipal wastewater using both individual elements, such as Fe, and multi-element otolith

signatures. Findings suggest that although certain elements may not always be useful for environmental reconstructions due to internal regulation, high levels of pollution can make differentiation possible, particularly for elements that are primarily associated with the salt fraction of otoliths. With further research, otolith microchemistry could evolve as an invaluable tool for tracing historical pollution in water bodies—particularly in regions that either lack baseline data or present monitoring challenges, such as the Arctic.

3.3 Conclusion

Otolith microchemistry has emerged as a powerful tool for tracing the life history of fish, providing insights into both physiological and environmental histories. This technique is especially valuable for understanding, monitoring, and managing Arctic ecosystems, which are sensitive to change but very poorly studied. In this study, I found that fish from a natural lagoon-tundra wastewater wetland exhibited multi-element otolith signatures. This difference was clear in both the initial year and the most recent periods of otolith growth. Additionally, I found that [Fe] may be useful in reconstructing contaminant histories in highly impacted systems. These results provided insights into the environmental conditions over time, surpassing what traditional soft tissue assessments can offer. I also observed that patterns of [Zn] and [Mn] in otolith cross-sections align with annuli, and that this association held true even when environmental concentrations were high, suggesting that chemical aging techniques may be useful even in contaminated environments.

Future research with finer temporal resolution could provide deeper insights into the link between environmental metal concentrations and otolith microchemistry. [Grønkjær \(2016\)](#) reported that plasma ion-protein complexes are key to understanding how various factors

influence otolith composition, making them a critical focus for further study. To clarify the mechanisms controlling otolith biomineralization and elemental incorporation, more comprehensive datasets linking otolith and environmental metal levels are essential. Additionally, integrating bioenergetic models that account for all processes affecting uptake and regulation will enhance our understanding ([Hüssy et al., 2024](#); [Reis-Santos et al., 2023](#); [Sturrock et al., 2015](#)). Overall, this research contributes to the expanding investigations of the utility of otolith microchemistry in environmental monitoring and assessment, and highlights areas for further, more comprehensive studies in the future.

Chapter 4

General Discussion

4.1 Research Synthesis

This thesis presents research on the impacts of municipal wastewater on Arctic ecosystems, specifically within the unique natural lagoon-tundra wastewater wetland in Baker Lake, the only inland community in Nunavut. My research explored trace metal dynamics across abiotic and biotic ecosystem compartments and investigated otolith microchemistry as a potential method to reconstruct wastewater exposure. Findings from this research not only filled knowledge gaps regarding the influence of municipal wastewater on cold, oligotrophic freshwater systems but also highlighted the potential use of otoliths as a marker of such exposures.

Results from the second chapter of this thesis revealed distinct differences in water chemistry between wastewater-affected lakes and reference lakes, primarily characterized by elevated concentrations of metals, including manganese (Mn), iron (Fe), and copper (Cu), as well as elevated concentrations of parameters that reflect primary productivity, such as chlorophyll-*a*. Despite observed improvements in water quality along the wastewater flow path, all wastewater-affected lakes had higher metal concentrations and primary productivity compared to reference lakes. Lagoon Lake, which receives wastewater directly from a stabilization pond, exhibited severely impacted water quality, with hypereutrophic conditions, high metal concentrations, and critically low dissolved oxygen levels that pose risks to aquatic life. Sediment analysis indicates elevated levels of Cu and zinc (Zn) near wastewater sources, although overall sediment chemistry showed no broad-scale differences between affected and reference sites. In terms of biotic impact, the influence of wastewater on Burbot (*Lota lota*) trophic ecology was

minimal, while Arctic grayling (*Thymallus arcticus*) showed enriched $\delta^{13}\text{C}$ in affected sites, suggesting changes in carbon sources. Muscle tissue analysis revealed lower (though not significant) methylmercury (MeHg) concentrations in fish from wastewater-affected areas compared to reference sites, despite higher MeHg concentrations in water, suggesting complex bioavailability and assimilation dynamics. These findings highlight the multifaceted impact of wastewater on aquatic ecosystems, and underscore need for ongoing monitoring and targeted management interventions.

In [Chapter 3](#) of this thesis, I examined hard part tissue chemistries of Arctic grayling from three study lakes, described as follows: downstream of influent, upstream of influent, and unimpacted. This investigation complemented previous analyses of abiotic environments and fish soft tissue chemistries.

Distinct multi-element otolith signatures (i.e., Zn, Mn, Cu, Fe, and Pb) among lakes were observed. Employing a multi-element approach may be beneficial in scenarios where trace metal concentrations are sub-lethal yet ecologically relevant, or when investigating exposure gradients, as they reflect increasingly subtle differences in otolith microchemistry ([Reis-Santos et al., 2018](#); [Sturrock et al., 2015](#)). Otoliths from a wastewater-impacted lake also exhibited significantly higher Fe concentrations in both early and recent growth stages compared to those from less impacted lakes, reflecting elevated [Fe] in water downstream of influent, as detailed in [Chapter 2](#). This suggests otoliths can record historical exposure to municipal wastewater, making them suitable for time-resolved environmental assessments when inferences are made carefully. Microchemical transects of otoliths from core to edge revealed clear minima and maxima of Zn

and Mn that aligned with annular structure, suggesting that chemical age estimates may be reliable even in contaminated environments.

Correlations between metal concentrations in otoliths and soft tissues (i.e., liver and muscle) were generally weak, potentially due to varying tissue metabolisms and time-lag effects of element deposition in otoliths ([Vignon et al., 2023](#)). Significant correlations were found between some transition metals in otoliths and fork length, age, and reproductive investment. Specifically, marginal edge Zn was inversely related to fish length across all lakes. This aligns with the role of transition metals (i.e., Zn, Mn, Cu, and Fe) as cofactors in protein-mediated crystallization processes (see [Thomas and Swearer, 2019](#)).

Results of [Chapter 3](#) enhance our understanding of otoliths for reconstructing physico-chemical histories, particularly with Fe, which is physiologically regulated and hypothesized not to reflect ambient water chemistry unless in heavily polluted settings ([Thomas et al., 2017](#); [Thomas and Swearer, 2019](#)). Here, results suggest that [Fe] may be useful as a tracer of effluent exposure, even in a field setting. By integrating otolith-based contaminant chronologies with other fish health indicators—such as soft tissue contaminant concentrations, trophic ecology, energy storage, and life history—results of this research could assist management and regulatory bodies in assessing the impacts of contaminants on fish and their ecosystems.

4.2 Limitations and Study Design

Otolith microchemistry reflects not only the external environment but also species-specific physiological processes, such as variability in metabolism, growth rates, and life histories. These biological factors can influence how elements are incorporated into otoliths, making it difficult to generalize findings across different species or even different populations of the same

species under varying environmental conditions. This work would have benefited from the inclusion of Burbot, especially since isotope ecology was found to differ from that of Arctic grayling ([Chapter 2](#)).

To effectively generalize across species, a deeper understanding of the mechanisms that govern incorporation of trace elements into the proteinaceous and calciferous matrices of otoliths is essential, along with insights into the species-specific protein-aragonite ratios. Research increasingly shows that the way elements are incorporated into otoliths is complex and non-linear, presenting challenges for using otolith microchemistry in reconstructions of past metal exposure. Elements that form divalent cations and substitute for calcium in the salt fractions of otoliths are more likely influenced by environmental conditions whereas elements essential to otolith matrix formation as protein co-factors are predominantly influenced by physiological processes ([Izzo et al., 2018](#); [Reis-Santos et al., 2023](#); [Thomas et al., 2020](#); [Thomas et al., 2017](#)). Elements, including Mn, that are present in both the salt and protein fractions likely reflect both environmental and physiological conditions, and further study of their use in reconstructing environmental histories is necessary, particularly as the factors influencing element incorporation may vary across different life stages ([Thomas et al., 2020](#)), and perhaps across gradients of environmental conditions. Multivariate statistical models that incorporate both environmental and biological covariates would also help disentangle the effects of external conditions from species-specific internal influences on otolith chemistry

This research is also limited by incomplete knowledge of fish movements within the Baker Lake wastewater treatment wetland. It is possible that Arctic grayling move up- and downstream of point source contamination at various life stages. Young-of-year (y.o.y.) Arctic grayling inhabit

the stream connecting Lagoon Lake to Upper Lagoon Lake ([Ellenor, 2020](#)), although it remains unclear if Lagoon Lake acts as a barrier to movement. Movement could introduce variability in otolith metal concentrations, complicating efforts to resolve contaminant histories.

Confounding variables, such as the proximity of the municipal landfill or roads, could also influence metal concentrations in the Baker Lake wastewater treatment wetland. Landfills, particularly older or poorly designed ones, can leach metals and other pollutants ([Slack et al., 2005](#)). Runoff from roads may carry heavy metals like lead, zinc, and copper from vehicle emissions and tire wear to downstream lakes (e.g., [Westerlund, 2007](#)). Additionally, dust from dirt or gravel roads can alter lake chemistry ([Zhu et al., 2019](#)) and biogeochemistry.

External contamination sources may have affected results by introducing metals into both the wastewater (from roads and landfill) and reference (from roads only) systems, complicating attribution of elevated metal concentrations in fish tissues solely to wastewater exposure. Future studies could quantify metal concentrations in soil and groundwater near, but unaffected by, the wetland. Additional reference sites further from pollution sources, along with regular monitoring of runoff and leachate from the landfill and roads, would help distinguish landfill-derived metals from those linked to wastewater.

4.3 Future work

This study could have been improved by: 1) investigating whether larval fish upstream and downstream of influents have unique nucleus metal concentrations and comparing these with adult fish otolith signatures; 2) implementing alternative methods for tracking fish movements, such as passive sampling (e.g., fyke nets) during and after the freshet; and 3) conducting finer-scale spatio-temporal analyses to identify specific locations and detect elevated

concentrations in metals, particularly following the annual freshet when the wastewater gravel berm is prone to failure. Despite this, the chosen scales of otolith and water chemistries were appropriate for addressing the research questions.

The Baker Lake community has expressed a desire for upgrades to the wastewater treatment wetland to enhance treatment efficacy. In anticipation of these improvements, further research could refine post-upgrade monitoring and assessment strategies. For instance, examining the otolith microchemistry of a second species with a limited home range, such as the small-bodied slimy sculpin (*Cottus cognatus*), could address limitations noted in [Chapter 3](#), allowing for a more comprehensive understanding of the abiotic and biotic impacts of wastewater discussed in [Chapter 2](#).

[4.4 Relevance to the Hamlet of Baker Lake and the wider Canadian Arctic](#)

This research is part of a larger program aimed at validating the environmental and human health benefits of upgrading passive municipal wastewater systems in Northern Canada. Concerns among community members about the proximity of final effluent discharge have persisted for over 25 years (documented in the Nunavut Impact Review Board public records). Upgrades in Baker Lake could mitigate these concerns by enabling the community to select a treatment option that respects traditional land use practices and considers their social, cultural, and economic needs. The work in this thesis has been actively shared with the community (see examples in [Figure S4.1](#) and [Figure S4.2](#)) and has fostered ongoing engagement through environmental sampling workshops involving local youth and members of the Baker Lake Hunters and Trappers Association.

In collaboration with the Hamlet of Baker Lake, Agnico-Eagle Mines Inc., a major industrial partner, has committed to developing a new passive wastewater treatment system. If realized, upgrades would support the community by improving environmental management practices and could offer the mine potential strategies for offsetting industry-related environmental impacts, which are regulatory requirements for its operations. Ultimately, the territorial government holds responsibility for initiating infrastructure upgrades within the community.

My thesis provides an initial characterization of the wastewater system before these upgrades, focusing on potential habitat improvements. One critical aspect is addressing the low dissolved oxygen levels in the lake immediately downstream from the influent, which, if improved, could serve as an effective strategy for habitat offsetting. Enhancing these conditions is vital for the survival and prosperity of species like the Arctic grayling, which are culturally and ecologically significant ([Hughson, 2010](#)).

If this approach proves successful, it could pave the way for implementing new, community-oriented offsetting projects in other northern communities, fostering sustainable environmental management that aligns with local needs and traditions. This could establish a model for community engagement and environmental stewardship across the Arctic.

4.5 Concluding Remarks

Overall, this work not only enhances scientific understanding but also provides practical tools and insights that can be used to improve environmental management, support development of future monitoring, and protect ecosystem and community health in the Arctic and similar regions worldwide.

References

- Abdi, H., Williams, L.J., 2010. Principal component analysis. *WIREs Comput. Stat.* 2, 433–459. <https://doi.org/10.1002/wics.101>
- Aguilar-Betancourt, C.M., González-Sansón, G., Kidd, K.A., Munkittrick, K.R., Curry, R.A., Kosonoy-Aceves, D., Lucano-Ramírez, G., Ruiz-Ramírez, S., Flores-Ortega, J.R., 2016. Fishes as indicators of untreated sewage contamination in a Mexican coastal lagoon. *Mar. Pollut. Bull.* 113, 100–109. <https://doi.org/10.1016/j.marpolbul.2016.08.073>
- Akaike, H., 1969. Fitting autoregressive models for prediction, in: Parzen, E., Tanabe, K., Kitagawa, G. (Eds.), *Selected Papers of Hirotugu Akaike*, Springer Series in Statistics. Springer New York, New York, NY, pp. 131–135. https://doi.org/10.1007/978-1-4612-1694-0_10
- Amundsen, P.-A., Bøhn, T., Popova, O.A., Staldvik, F.J., Reshetnikov, Y.S., Kashulin, N.A., Lukin, A.A., 2003. Ontogenetic niche shifts and resource partitioning in a subarctic piscivore fish guild. *Hydrobiologia* 497, 109–119. <https://doi.org/10.1023/A:1025465705717>
- Amundsen, P.-A., Henriksson, M., Poste, A., Prati, S., Power, M., 2023. Ecological drivers of mercury bioaccumulation in fish of a Subarctic watercourse. *Environ. Toxicol. Chem.* 42, 873–887. <https://doi.org/10.1002/etc.5580>
- Anderson, M.J., 2001. A new method for non-parametric multivariate analysis of variance. *Austral Ecol.* 26, 32–46. <https://doi.org/10.1111/j.1442-9993.2001.01070.pp.x>
- Back, D.-Y., Ha, S.-Y., Else, B., Hanson, M., Jones, S.F., Shin, K.-H., Tatarek, A., Wiktor, J.M., Cicek, N., Alam, S., Mundy, C.J., 2021. On the impact of wastewater effluent on phytoplankton in the Arctic coastal zone: A case study in the Kitikmeot Sea of the Canadian Arctic. *Sci. Total Environ.* 764, 143861. <https://doi.org/10.1016/j.scitotenv.2020.143861>
- Balch, G., Hayward, J., Jamieson, R., Wootton, B., Yates, C.N., 2018. Recommendations for the use of tundra wetlands for treatment of municipal wastewater in Canada's far North, in: Nagabhatla, N., Metcalfe, C.D. (Eds.), *Multifunctional Wetlands, Environmental Contamination Remediation and Management*. Springer International Publishing, pp. 83–120. https://doi.org/10.1007/978-3-319-67416-2_3
- Barnes, T.C., Gillanders, B.M., 2013. Combined effects of extrinsic and intrinsic factors on otolith chemistry: implications for environmental reconstructions. *Can. J. Fish. Aquat. Sci.* 70, 1159–1166. <https://doi.org/10.1139/cjfas-2012-0442>
- Barr, D.J., Levy, R., Scheepers, C., Tily, H.J., 2013. Random effects structure for confirmatory hypothesis testing: Keep it maximal. *J. Mem. Lang.* 68, 10.1016/j.jml.2012.11.001.
- Bartlett, M.S., 1937. Properties of sufficiency and statistical tests. *Proc. R. Soc. Lond. Ser. - Math. Phys. Sci.* 160, 268–282. <https://doi.org/10.1098/rspa.1937.0109>
- Bates, D., Maechler, M., Bolker, B., Walker, S., Christensen, R.H.B., Singmann, H., Dai, B., Scheipl, F., Grothendieck, G., Green, P., 2009. Package 'lme4.' [10.32614/CRAN.package.lme4](https://doi.org/10.32614/CRAN.package.lme4)
- Birtwell, I.K., 1984. A brief investigation of Arctic grayling (*Thymallus arcticus*) and aquatic invertebrates in the Minto Creek drainage, Mayo, Yukon Territory: an area subjected to placer mining. Department of Fisheries and Oceans, Fisheries Research Branch, West

- Vancouver Branch. https://publications.gc.ca/collections/collection_2021/mpo-dfo/Fs97-6-1287-eng.pdf
- Brown, C.J., Knight, B.W., McMaster, M.E., Munkittrick, K.R., Oakes, K.D., Tetreault, G.R., Servos, M.R., 2011. The effects of tertiary treated municipal wastewater on fish communities of a small river tributary in Southern Ontario, Canada. *Environ. Pollut.* 159, 1923–1931. <https://doi.org/10.1016/j.envpol.2011.03.014>
- Bryan, G.W., 1976. Some aspects of heavy metal tolerance in aquatic organisms. *Eff. Pollut. Aquat. Org.* Cambridge University Press, Cambridge p. 7-34.
- Cabana, G., Rasmussen, J.B., 1996. Comparison of aquatic food chains using nitrogen isotopes. *Proc. Natl. Acad. Sci.* 93, 10844–10847. <https://doi.org/10.1073/pnas.93.20.10844>
- Cabana, G., Rasmussen, J.B., 1994. Modelling food chain structure and contaminant bioaccumulation using stable nitrogen isotopes. *Nature* 372, 255–257. <https://doi.org/10.1038/372255a0>
- Cabana, G., Tremblay, A., Kalff, J., Rasmussen, J.B., 1994. Pelagic food chain structure in Ontario lakes: a determinant of mercury levels in Lake trout (*Salvelinus namaycush*). *Can. J. Fish. Aquat. Sci.* 51, 381–389. <https://doi.org/10.1139/f94-039>
- Campana, S.E., 2005. Otolith elemental composition as a natural marker of fish stocks, in: *Stock Identification Methods*. Elsevier, pp. 227–245. <https://doi.org/10.1016/B978-012154351-8/50013-7>
- Campana, S.E., 1999. Chemistry and composition of fish otoliths: pathways, mechanisms and applications. *Mar. Ecol. Prog. Ser.* 188, 263–297. <https://doi.org/10.3354/meps188263>
- Campana, S.E., 1983. Feeding periodicity and the production of daily growth increments in otoliths of Steelhead trout (*Salmo gairdneri*) and Starry flounder (*Platichthys stellatus*). *Can. J. Zool.* 61, 1591–1597. <https://doi.org/10.1139/z83-214>
- Campana, S.E., Fowler, A.J., Jones, C.M., 1994. Otolith elemental fingerprinting for stock identification of Atlantic Cod (*Gadus morhua*) using laser ablation ICPMS. *Can. J. Fish. Aquat. Sci.* 51, 1942–1950. <https://doi.org/10.1139/f94-196>
- Campana, S.E., Neilson, J.D., 1985. Microstructure of fish otoliths. *Can. J. Fish. Aquat. Sci.* 42, 1014–1032. <https://doi.org/10.1139/f85-127>
- Campbell, L.M., Norstrom, R.J., Hobson, K.A., Muir, D.C.G., Backus, S., Fisk, A.T., 2005. Mercury and other trace elements in a pelagic Arctic marine food web (North water Polynya, Baffin Bay). *Sci. Total Environ.* 351–352, 247–263. <https://doi.org/10.1016/j.scitotenv.2005.02.043>
- Carlson, R.E., 1977. A trophic state index for lakes. *Limnol. Oceanogr.* 22, 361–369. <https://doi.org/10.4319/lo.1977.22.2.0361>
- Casselman, J.M., 1983. Age and growth assessment of fish from their calcified structures—techniques and tools. NOAA Tech. Rep. NMFS 8, 1–17. <https://spo.nmfs.noaa.gov/Technical%20Report/tr8opt.pdf>
- CCME, 2001. Canadian water quality guidelines for the protection of aquatic life: CCME water quality index 1.0. <https://ccme.ca/en/res/wqimanualen.pdf>
- Chambers, P.A., Allard, M., Walker, S.L., Marsalek, J., Lawrence, J., Servos, M., Busnarda, J., Munger, K.S., Adare, K., Jefferson, C., Kent, R.A., Wong, M.P., 1997. Impacts of municipal wastewater effluents on Canadian waters: a review. *Water Qual. Res. J.* 32, 659–714. <https://doi.org/10.2166/wqrj.1997.038>

- Chan, Y.J., Chong, M.F., Law, C.L., Hassell, D.G., 2009. A review on anaerobic–aerobic treatment of industrial and municipal wastewater. *Chem. Eng. J.* 155, 1–18.
<https://doi.org/10.1016/j.cej.2009.06.041>
- Chandra, S., Jake Vander Zanden, M., Heyvaert, A.C., Richards, B.C., Allen, B.C., Goldman, C.R., 2005. The effects of cultural eutrophication on the coupling between pelagic primary producers and benthic consumers. *Limnol. Oceanogr.* 50, 1368–1376.
<https://doi.org/10.4319/lo.2005.50.5.1368>
- Chandrapalan, T., Kwong, R.W.M., 2021. Functional significance and physiological regulation of essential trace metals in fish. *J. Exp. Biol.* 224, jeb238790.
<https://doi.org/10.1242/jeb.238790>
- Chaves-Barquero, L.G., Luong, K.H., Mundy, C.J., Knapp, C.W., Hanson, M.L., Wong, C.S., 2016. The release of wastewater contaminants in the Arctic: A case study from Cambridge Bay, Nunavut, Canada. *Environ. Pollut.* 218, 542–550.
<https://doi.org/10.1016/j.envpol.2016.07.036>
- Chételat, J., Amyot, M., Garcia, E., 2011. Habitat-specific bioaccumulation of methylmercury in invertebrates of small mid-latitude lakes in North America. *Environ. Pollut.* 159, 10–17.
<https://doi.org/10.1016/j.envpol.2010.09.034>
- Chipasa, K.B., K.B., 2003. Accumulation and fate of selected heavy metals in a biological wastewater treatment system. *Waste Manag.* 23, 135–143.
[https://doi.org/10.1016/S0956-053X\(02\)00065-X](https://doi.org/10.1016/S0956-053X(02)00065-X)
- Chouinard, A., Yates, C.N., Balch, G.C., Jørgensen, S.E., Wootton, B.C., Anderson, B.C., 2014. Management of tundra wastewater treatment wetlands within a lagoon/wetland hybridized treatment system using the SubWet 2.0 Wetland Model. *Water* 6, 439–454.
<https://doi.org/10.3390/w6030439>
- Cott, P.A., Johnston, T.A., Gunn, J.M., 2011. Food web position of burbot relative to lake trout, northern pike, and lake whitefish in four sub-Arctic boreal lakes. *J. Appl. Ichthyol.* 27, 49–56. <https://doi.org/10.1111/j.1439-0426.2011.01843.x>
- Daley, K., Castleden, H., Jamieson, R., Furgal, C., Ell, L., 2015. Water systems, sanitation, and public health risks in remote communities: Inuit resident perspectives from the Canadian Arctic. *Soc. Sci. Med.* 135, 124–132.
<https://doi.org/10.1016/j.socscimed.2015.04.017>
- Daley, K., Jamieson, R., Rainham, D., Hansen, L.T., Harper, S.L., 2022. Microbial risk assessment and mitigation options for wastewater treatment in Arctic Canada. *Microb. Risk Anal.* 20, 100186. <https://doi.org/10.1016/j.mran.2021.100186>
- Daley, K., Jamieson, R., Rainham, D., Truelstrup Hansen, L., 2018. Wastewater treatment and public health in Nunavut: a microbial risk assessment framework for the Canadian Arctic. *Environ. Sci. Pollut. Res.* 25, 32860–32872. <https://doi.org/10.1007/s11356-017-8566-8>
- Daley, K., Jamieson, R., Rainham, D., Truelstrup Hansen, L., Harper, S.L., 2019. Screening-level microbial risk assessment of acute gastrointestinal illness attributable to wastewater treatment systems in Nunavut, Canada. *Sci. Total Environ.* 657, 1253–1264.
<https://doi.org/10.1016/j.scitotenv.2018.11.408>

- Daskin, J.H., Calci, K.R., Burkhardt, W., Carmichael, R.H., 2008. Use of N stable isotope and microbial analyses to define wastewater influence in Mobile Bay, AL. *Mar. Pollut. Bull.* 56, 860–868. <https://doi.org/10.1016/j.marpolbul.2008.02.002>
- Dawson, R.N., Grainge, J.W., 1969. Proposed design criteria for wastewater lagoons in Arctic and Sub-Arctic regions. *J. Water Pollut. Control Fed.* 41, 237–246. <https://www.jstor.org/stable/25036238>
- deBruyn, A.M.H., Rasmussen, J.B., 2002. Quantifying Assimilation of Sewage-Derived Organic Matter by Riverine Benthos. *Ecol. Appl.* 12, 511–520. [https://doi.org/10.1890/1051-0761\(2002\)012\[0511:QAOSDO\]2.0.CO;2](https://doi.org/10.1890/1051-0761(2002)012[0511:QAOSDO]2.0.CO;2)
- Denoyer, E.R., Fredeen, K.J., Hager, J.W., 1991. Laser Solid Sampling for Inductively Coupled Plasma Mass Spectrometry. *Anal. Chem.* 63, 445A-457A. <https://doi.org/10.1021/ac00008a718>
- Di Toro, D.M., Allen, H.E., Bergman, H.L., Meyer, J.S., Paquin, P.R., Santore, R.C., 2001. Biotic ligand model of the acute toxicity of metals. 1. Technical Basis. *Environ. Toxicol. Chem.* 20, 2383–2396. <https://doi.org/10.1002/etc.5620201034>
- Dorval, E., Jones, C.M., Hannigan, R., Van Montfrans, J., 2007. Relating otolith chemistry to surface water chemistry in a coastal plain estuary. *Can. J. Fish. Aquat. Sci.* 64, 411–424. <https://doi.org/10.1139/F07-015>
- Driscoll, C.T., Mason, R.P., Chan, H.M., Jacob, D.J., Pirrone, N., 2013. Mercury as a global pollutant: sources, pathways, and effects. *Environ. Sci. Technol.* 47, 4967–4983. <https://doi.org/10.1021/es305071v>
- Dunbabin, J.S., Bowmer, K.H., 1992. Potential use of constructed wetlands for treatment of industrial wastewaters containing metals. *Sci. Total Environ.* 111, 151–168. [https://doi.org/10.1016/0048-9697\(92\)90353-T](https://doi.org/10.1016/0048-9697(92)90353-T)
- ECCC, 2021. Federal environmental quality guidelines for copper. <https://www.canada.ca/content/dam/eccc/documents/pdf/pded/feqg-copper/Federal-Environmental-Quality-Guidelines-Copper.pdf>
- ECCC, 2015. Wastewater Systems Effluent Regulations. <https://laws-lois.justice.gc.ca/eng/regulations/sor-2012-139/FullText.html>
- Ellenor, J., 2020. Habitat use of young-of-year Arctic grayling (*Thymallus arcticus*) in Barrenland streams of central Nunavut, Canada (Master's Thesis). University of Waterloo, Waterloo, ON.
- Elsdon, T.S., Gillanders, B.M., 2002. Interactive effects of temperature and salinity on otolith chemistry: Challenges for determining environmental histories of fish. *Can. J. Fish. Aquat. Sci.* 59, 1796–1808. <https://doi.org/10.1139/f02-154>
- Elsdon, T.S., Wells, B.K., Campana, S.E., Gillanders, B.M., Jones, C.M., Limburg, K.E., Secor, D.H., Thorrold, S.R., Walther, B.D., 2008. Otolith chemistry to describe movements and life-history parameters of fishes: hypotheses, assumptions, limitations and inferences. *Oceanogr. Mar. Biol. Annu. Rev.* 46, 297–330. <http://dx.doi.org/10.3354/meps14646>
- EMAN-North, 2005. Northern waters: a guide to designing and conducting water quality monitoring in Northern Canada. Northern Ecological Monitoring Assessment Network. https://publications.gc.ca/collections/collection_2014/ec/En84-113-2005-eng.pdf
- Evans, M.S., Lockhart, W.L., Doetzel, L., Low, G., Muir, D., Kidd, K., Stephens, G., Delaronde, J., 2005. Elevated mercury concentrations in fish in lakes in the Mackenzie River Basin: The

- role of physical, chemical, and biological factors. *Sci. Total Environ.* 351–352, 479–500. <https://doi.org/10.1016/j.scitotenv.2004.12.086>
- Farkas, A., Salánki, J., Specziár, A., 2002. Relation between growth and the heavy metal concentration in organs of Bream *Abramis brama* L. populating Lake Balaton. *Arch. Environ. Contam. Toxicol.* 43, 236–243. <https://doi.org/10.1007/s00244-002-1123-5>
- Focken, U., Becker, K., 1998. Metabolic fractionation of stable carbon isotopes: implications of different proximate compositions for studies of the aquatic food webs using $\delta^{13}\text{C}$ data. *Oecologia* 115, 337–343. <https://doi.org/10.1007/s004420050525>
- France, R.L., 1995. Differentiation between littoral and pelagic food webs in lakes using stable carbon isotopes. *Limnol. Oceanogr.* 40, 1310–1313. <https://doi.org/10.4319/lo.1995.40.7.1310>
- Fredrickson, J.K., Gorby, Y.A., 1996. Environmental processes mediated by iron-reducing bacteria. *Curr. Opin. Biotechnol.* 7, 287–294. [https://doi.org/10.1016/S0958-1669\(96\)80032-2](https://doi.org/10.1016/S0958-1669(96)80032-2)
- Friedrich, L.A., Halden, N.M., 2010. Determining exposure history of Northern pike and Walleye to tailings effluence using trace metal uptake in otoliths. *Environ. Sci. Technol.* 44, 1551–1558. <https://doi.org/10.1021/es903261q>
- Froese, R., 2006. Cube law, condition factor and weight–length relationships: history, meta-analysis and recommendations. *J. Appl. Ichthyol.* 22, 241–253. <https://doi.org/10.1111/j.1439-0426.2006.00805.x>
- Fuzzen, M.L.M., Bragg, L.M., Tetreault, G.R., Bahamonde, P.A., Tanna, R.N., Bennett, C.J., McMaster, M.E., Servos, M.R., 2016. An assessment of the spatial and temporal variability of biological responses to municipal wastewater effluent in Rainbow darter (*Etheostoma caeruleum*) collected along an urban gradient. *PLoS ONE* 11, e0164879. <https://doi.org/10.1371/journal.pone.0164879>
- Gartner, A., Lavery, P., Smit, A.J., 2002. Use of $\delta^{15}\text{N}$ signatures of different functional forms of macroalgae and filter-feeders to reveal temporal and spatial patterns in sewage dispersal. *Mar. Ecol. Prog. Ser.* 235, 63–73. <https://doi.org/10.3354/meps235063>
- Gaston, T.F., Suthers, I.M., 2004. Spatial variation in $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ of liver, muscle and bone in a rocky reef planktivorous fish: the relative contribution of sewage. *J. Exp. Mar. Biol. Ecol.* 304, 17–33. <https://doi.org/10.1016/j.jembe.2003.11.022>
- Gibbons, W.N., Munkittrick, K.R., 1994. A sentinel monitoring framework for identifying fish population responses to industrial discharges. *J. Aquat. Ecosyst. Health* 3, 227–237. <https://doi.org/10.1007/BF00043244>
- Goedkoop, W., Culp, J.M., Christensen, T., Christoffersen, K.S., Fefilova, E., Guðbergsson, G., Lárusson, K.F., Liljaniemi, P., Novichkova, A.A., Ólafsson, J.S., Sandøy, S., Lento, J., 2022. Improving the framework for assessment of ecological change in the Arctic: A circumpolar synthesis of freshwater biodiversity. *Freshw. Biol.* 67, 210–223. <https://doi.org/10.1111/fwb.13873>
- Gotelli, N.J., Ellison, A.M., 2013. A primer of ecological statistics, second edition. ed. Sinauer Associates Inc., Sunderland, Massachusetts, USA.
- Gower, J.C., 1966. Some distance properties of latent root and vector methods used in multivariate analysis. *Biometrika* 53, 325–338. <https://doi.org/10.1093/biomet/53.3-4.325>

- Grammer, G.L., Morrongiello, J.R., Izzo, C., Hawthorne, P.J., Middleton, J.F., Gillanders, B.M., 2017. Coupling biogeochemical tracers with fish growth reveals physiological and environmental controls on otolith chemistry. *Ecol. Monogr.* 87, 487–507. <https://doi.org/10.1002/ecm.1264>
- Gromala, M., Neufeld, J.D., McConkey, B.J., 2021. Monitoring microbial populations and antibiotic resistance gene enrichment associated with Arctic waste stabilization ponds. *Appl. Environ. Microbiol.* 87, e02914-20. <https://doi.org/10.1128/AEM.02914-20>
- Grønkjær, P., 2016. Otoliths as individual indicators: a reappraisal of the link between fish physiology and otolith characteristics. *Mar. Freshw. Res.* 67, 881–888. <https://doi.org/10.1071/MF15155>
- Halden, N.M., Mejia, S.R., Babaluk, J.A., Reist, J.D., Kristofferson, A.H., Campbell, J.L., Teesdale, W.J., 2000. Oscillatory zinc distribution in Arctic char (*Salvelinus alpinus*) otoliths: the result of biology or environment? *Fish. Res.* 46, 289–298. [https://doi.org/10.1016/S0165-7836\(00\)00154-5](https://doi.org/10.1016/S0165-7836(00)00154-5)
- Hansson, S.V., Desforges, J.-P., van Beest, F.M., Bach, L., Halden, N.M., Sonne, C., Mosbech, A., Søndergaard, J., 2020. Bioaccumulation of mining derived metals in blood, liver, muscle and otoliths of two Arctic predatory fish species (*Gadus ogac* and *Myoxocephalus scorpius*). *Environ. Res.* 183, 109194. <https://doi.org/10.1016/j.envres.2020.109194>
- Harrell Jr, F.E., 2019. Package ‘hmisc.’ CRAN2018 2019, 235–236. <https://cran.r-project.org/web/packages/Hmisc/Hmisc.pdf>
- Harrison, X.A., Donaldson, L., Correa-Cano, M.E., Evans, J., Fisher, D.N., Goodwin, C.E.D., Robinson, B.S., Hodgson, D.J., Inger, R., 2018. A brief introduction to mixed effects modelling and multi-model inference in ecology. *PeerJ*, 6, e4794. <https://doi.org/10.7717/peerj.4794>
- Hayden, B., Harrod, C., Thomas, S.M., Eloranta, A.P., Myllykangas, J.-P., Siwertsson, A., Præbel, K., Knudsen, R., Amundsen, P.-A., Kahilainen, K.K., 2019. From clear lakes to murky waters – tracing the functional response of high-latitude lake communities to concurrent ‘greening’ and ‘browning.’ *Ecol. Lett.* 22, 807–816. <https://doi.org/10.1111/ele.13238>
- Hayward, J.L., Jamieson, R., Boutilier, L., Goulden, T., Lam, B., 2014. Treatment performance assessment and hydrological characterization of an arctic tundra wetland receiving primary treated municipal wastewater. *Ecol. Eng.* 73, 786–797. <https://doi.org/10.1016/j.ecoleng.2014.09.107>
- Hayward, J.L., Jackson, A.J., Yost, C.K., Hansen, L., Jamieson, R.C., 2018. Fate of antibiotic resistance genes in two Arctic tundra wetlands impacted by municipal wastewater. *Sci. Total Environ.* 642, 1415–1428. <https://doi.org/10.1016/j.scitotenv.2018.06.083>
- Health Canada, 2010. Federal contaminated site risk assessment in Canada : supplemental guidance on human health risk assessment for country foods (HHRA Foods) / prepared by Contaminated Sites Division, Safe Environments Directorate. Ottawa, Ontario. https://publications.gc.ca/collections/collection_2012/sc-hc/H128-1-11-641-eng.pdf
- Health Canada, 2007. Federal Environmental Quality Guidelines (FEQGs). Accessed 4.16.24. <https://www.canada.ca/en/health-canada/services/chemical-substances/fact-sheets/federal-environmental-quality-guidelines.html>

- Heath, A.G., 1995. Water pollution and fish physiology. CRC Press.
<https://doi.org/10.1201/9780203718896>
- Heaton, T.H., 1986. Isotopic studies of nitrogen pollution in the hydrosphere and atmosphere: a review. *Chem. Geol. Isot. Geosci. Sect.* 59, 87–102. [https://doi.org/10.1016/0168-9622\(86\)90059-X](https://doi.org/10.1016/0168-9622(86)90059-X)
- Hebbali, A., 2020. *olsrr: Tools for building OLS regression models*. R Package Version 05 3.
- Hecky, R.E., Hesslein, R.H., 1995. Contributions of benthic algae to lake food webs as revealed by stable isotope analysis. *J. North Am. Benthol. Soc.* 14, 631–653.
<https://doi.org/10.2307/1467546>
- Heimbrand, Y., Limburg, K.E., Hüsey, K., Casini, M., Sjöberg, R., Palmén Bratt, A.-M., Levinsky, S.-E., Karpushevskaya, A., Radtke, K., Öhlund, J., 2020. Seeking the true time: exploring otolith chemistry as an age-determination tool. *J. Fish Biol.* 97, 552–565.
<https://doi.org/10.1111/jfb.14422>
- Heino, J., Culp, J.M., Erkinaro, J., Goedkoop, W., Lento, J., Rühland, K.M., Smol, J.P., 2020. Abruptly and irreversibly changing Arctic freshwaters urgently require standardized monitoring. *J. Appl. Ecol.* 57, 1192–1198. <https://doi.org/10.1111/1365-2664.13645>
- Hicks, K.A., Loomer, H.A., Fuzzen, M.L.M., Kleywegt, S., Tetreault, G.R., McMaster, M.E., Servos, M.R., 2017. $\delta^{15}\text{N}$ tracks changes in the assimilation of sewage-derived nutrients into a riverine food web before and after major process alterations at two municipal wastewater treatment plants. *Ecol. Indic.* 72, 747–758.
<https://doi.org/10.1016/j.ecolind.2016.09.011>
- Holeton, C., Chambers, P.A., Grace, L., 2011. Wastewater release and its impacts on Canadian waters. *Can. J. Fish. Aquat. Sci.* 68, 1836–1859. <https://doi.org/10.1139/f2011-096>
- Huang, Y., Ragush, C.M., Johnston, L.H., Hall, M.W., Beiko, R.G., Jamieson, R.C., Hansen, L., 2021. Changes in bacterial communities during treatment of municipal wastewater in Arctic wastewater stabilization ponds. *Front. Water* 3, 710853.
<https://doi.org/10.3389/frwa.2021.710853>
- Hughson, P.K., 2010. Our homeland for the past, present and future: Akulliqpaaq Qamaniq (Aberdeen Lake) and Qamaniq Tugliqpaq (Schultz Lake) landscapes described by Elder John Killulark. <http://hdl.handle.net/1993/4115>
- Huser, B.J., Futter, M.N., Bogan, D., Brittain, J.E., Culp, J.M., Goedkoop, W., Gribovskaya, I., Karlsson, J., Lau, D.C.P., Rühland, K.M., Schartau, A.K., Shaftel, R., Smol, J.P., Vrede, T., Lento, J., 2022. Spatial and temporal variation in Arctic freshwater chemistry—reflecting climate-induced landscape alterations and a changing template for biodiversity. *Freshw. Biol.* 67, 14–29. <https://doi.org/10.1111/fwb.13645>
- Hüsey, K., 2008. Otolith accretion rates: Does size really matter? *J. Exp. Mar. Biol. Ecol.* 362, 131–136. <https://doi.org/10.1016/j.jembe.2008.06.016>
- Hüsey, K., Haase, S., Mion, M., Hilvarsson, A., Radtke, K., Thomsen, T.B., Krüger-Johnsen, M., Casini, M., Sturrock, A.M., 2024. Into the wild: coupling otolith and archival tag records to test assumptions underpinning otolith chemistry applications in wild fish. *Front. Mar. Sci.* 11. <https://doi.org/10.3389/fmars.2024.1365023>
- Hüsey, K., Krüger-Johnsen, M., Thomsen, T.B., Heredia, B.D., Næraa, T., Limburg, K.E., Heimbrand, Y., McQueen, K., Haase, S., Krumme, U., Casini, M., Mion, M., Radtke, K., 2021a. It's elemental, my dear Watson: validating seasonal patterns in otolith chemical

- chronologies. *Can. J. Fish. Aquat. Sci.* 78, 551–566. <https://doi.org/10.1139/cjfas-2020-0388>
- Hüssy, K., Limburg, K.E., de Pontual, H., Thomas, O.R.B., Cook, P.K., Heimbrand, Y., Blass, M., Sturrock, A.M., 2021b. Trace element patterns in otoliths: the role of biomineralization. *Rev. Fish. Sci. Aquac.* 29, 445–477. <https://doi.org/10.1080/23308249.2020.1760204>
- Izzo, C., Doubleday, Z.A., Gillanders, B.M., Izzo, C., Doubleday, Z.A., Gillanders, B.M., 2015. Where do elements bind within the otoliths of fish? *Mar. Freshw. Res.* 67, 1072–1076. <https://doi.org/10.1071/MF15064>
- Izzo, C., Reis-Santos, P., Gillanders, B.M., 2018. Otolith chemistry does not just reflect environmental conditions: a meta-analytic evaluation. *Fish Fish.* 19, 441–454. <https://doi.org/10.1111/faf.12264>
- Johnson, K., 2008. National Inuit position paper regarding the CCME Canada-Wide strategy for the management of municipal wastewater effluent and environment Canada's proposed regulatory framework for wastewater. Inuit Tapiriit Kanatami, Ottawa. <https://www.itk.ca/wp-content/uploads/2016/07/position-paper-regarding.pdf>
- Kalish, J.M., 1989. Otolith microchemistry: validation of the effects of physiology, age and environment on otolith composition. *J. Exp. Mar. Biol. Ecol.* 132, 151–178. [https://doi.org/10.1016/0022-0981\(89\)90126-3](https://doi.org/10.1016/0022-0981(89)90126-3)
- Karimi, R., Chen, C.Y., Folt, C.L., 2016. Comparing nearshore benthic and pelagic prey as mercury sources to lake fish: the importance of prey quality and mercury content. *Sci. Total Environ.* 565, 211–221. <https://doi.org/10.1016/j.scitotenv.2016.04.162>
- Karimi, R., Chen, C.Y., Pickhardt, P.C., Fisher, N.S., Folt, C.L., 2007. Stoichiometric controls of mercury dilution by growth. *Proc. Natl. Acad. Sci.* 104, 7477–7482. <https://doi.org/10.1073/pnas.0611261104>
- Kassambara, A., 2016. Factoextra: extract and visualize the results of multivariate data analyses. R Package Version 1. [10.32614/CRAN.package.factoextra](https://cran.r-project.org/web/packages/factoextra/index.html)
- Kennedy, B.P., Klaue, A., Blum, J.D., Folt, C.L., Nislow, K.H., 2002. Reconstructing the lives of fish using Sr isotopes in otoliths. *Can. J. Fish. Aquat. Sci.* 59, 925–929. <https://doi.org/10.1139/f02-070>
- Kidd, K.A., Hesslein, R.H., Fudge, R.J.P., Hallard, K.A., 1995a. The influence of trophic level as measured by $\delta^{15}\text{N}$ on mercury concentrations in freshwater organisms. *Water. Air. Soil Pollut.* 80, 1011–1015. <https://doi.org/10.1007/BF01189756>
- Kidd, K.A., Muir, D.C.G., Evans, M.S., Wang, X., Whittle, M., Swanson, H.K., Johnston, T., Guildford, S., 2012. Biomagnification of mercury through Lake trout (*Salvelinus namaycush*) food webs of lakes with different physical, chemical and biological characteristics. *Sci. Total Environ.* 438, 135–143. <https://doi.org/10.1016/j.scitotenv.2012.08.057>
- Kidd, K.A., Schindler, D.W., Hesslein, R.H., Muir, D.C.G., 1995b. Correlation between stable nitrogen isotope ratios and concentrations of organochlorines in biota from a freshwater food web. *Sci. Total Environ.* 160, 381–390. [https://doi.org/10.1016/0048-9697\(95\)04371-7](https://doi.org/10.1016/0048-9697(95)04371-7)
- Kigjugalik Webster, D., 2001. Harvaqtuurmiut heritage: the heritage of the Inuit of the Lower Kazan River. Inuit Heritage Centre, Baker Lake, Nunavut. Artisan Press, Yellowknife.

- Kilgour, B.W., Dubé, M.G., Hedley, K., Portt, C.B., Munkittrick, K.R., 2007. Aquatic environmental effects monitoring guidance for environmental assessment practitioners. *Environ. Monit. Assess.* 130, 423–436. <https://doi.org/10.1007/s10661-006-9433-0>
- Kilgour, B.W., Munkittrick, K.R., Portt, C.B., Hedley, K., Culp, J., Dixit, S., Pastershank, G., 2005. Biological criteria for municipal wastewater effluent monitoring programs. *Water Qual. Res. J.* 40, 374–387. <https://doi.org/10.2166/wqrj.2005.041>
- Kitikmeot Inuit Association, 2006. Volume II fish and wildlife: Ekalok (fish) and fishing (No. 1), Naonaiyaotit Traditional Knowledge Project. Cambridge Bay and Kugluktuk, NU.
- Kraemer, L.D., Campbell, P.G.C., Hare, L., 2005. A field study examining metal elimination kinetics in juvenile Yellow perch (*Perca flavescens*). *Aquat. Toxicol.* 75, 108–126. <https://doi.org/10.1016/j.aquatox.2005.07.007>
- Kroll, C.N., Song, P., 2013. Impact of multicollinearity on small sample hydrologic regression models. *Water Resour. Res.* 49, 3756–3769. <https://doi.org/10.1002/wrcr.20315>
- Krumhansl, K.A., Krkosek, W.H., Greenwood, M., Ragush, C., Schmidt, J., Grant, J., Barrell, J., Lu, L., Lam, B., Gagnon, G.A., Jamieson, R.C., 2015. Assessment of Arctic community wastewater impacts on marine benthic invertebrates. *Environ. Sci. Technol.* 49, 760–766. <https://doi.org/10.1021/es503330n>
- Lam, B., Livingston, T., 2011. Active research into passive systems: a study of wastewater in Nunavut. *J. North. Territ. Water Waste Assoc.* 7, 42–45. https://ntwwa.com/wp-content/uploads/2020/05/Journal_2011_Web.pdf
- Laske, S.M., Rosenberger, A.E., Wipfli, M.S., Zimmerman, C.E., 2018. Generalist feeding strategies in Arctic freshwater fish: a mechanism for dealing with extreme environments. *Ecol. Freshw. Fish* 27, 767–784. <https://doi.org/10.1111/eff.12391>
- Lavoie, R.A., Jardine, T.D., Chumchal, M.M., Kidd, K.A., Campbell, L.M., 2013. Biomagnification of mercury in aquatic food webs: a worldwide meta-analysis. *Environ. Sci. Technol.* 47, 13385–13394. <https://doi.org/10.1021/es403103t>
- Le Cren, E.D., 1951. The length-weight relationship and seasonal cycle in gonad weight and condition in the Perch (*Perca fluviatilis*). *J. Anim. Ecol.* 20, 201–219. <https://doi.org/10.2307/1540>
- Lesage, E., Rousseau, D.P.L., Meers, E., Tack, F.M.G., De Pauw, N., 2007a. Accumulation of metals in a horizontal subsurface flow constructed wetland treating domestic wastewater in Flanders, Belgium. *Sci. Total Environ.* 380, 102–115. <https://doi.org/10.1016/j.scitotenv.2006.10.055>
- Lesage, E., Rousseau, D.P.L., Meers, E., Van de Moortel, A.M.K., Du Laing, G., Tack, F.M.G., De Pauw, N., Verloo, M.G., 2007b. Accumulation of metals in the sediment and reed biomass of a combined constructed wetland treating domestic wastewater. *Water. Air. Soil Pollut.* 183, 253–264. <https://doi.org/10.1007/s11270-007-9374-4>
- Lescord, G.L., Johnston, T.A., Branfireun, B.A., Gunn, J.M., 2018. Percentage of methylmercury in the muscle tissue of freshwater fish varies with body size and age and among species. *Environ. Toxicol. Chem.* 37, 2682–2691. <https://doi.org/10.1002/etc.4233>
- Limburg, K.E., Walther, B.D., Lu, Z., Jackman, G., Mohan, J., Walther, Y., Nissling, A., Weber, P.K., Schmitt, A.K., 2015. In search of the dead zone: use of otoliths for tracking fish exposure to hypoxia. *J. Mar. Syst.* 141, 167–178. <https://doi.org/10.1016/j.jmarsys.2014.02.014>

- Loomer, H.A., Oakes, K.D., Schiff, S.L., Taylor, W.D., Servos, M.R., 2015. Use of Stable Isotopes to Trace Municipal Wastewater Effluents into Food Webs within a Highly Developed River System. *River Res. Appl.* 31, 1093–1100. <https://doi.org/10.1002/rra.2826>
- McMaster, M.E., Hewitt, L.M., Tetreault, G.R., Janoscik, T., Boyko, C., Peters, L., Parrott, J.L., Van Der Kraak, G.J., Portt, C.B., Kroll, K.J., 2005. Detailed endocrine assessments of wild fish in the Northern River Basins, Alberta, in comparison to EEM monitored endpoints. *Water Qual. Res. J.* 40, 299–314. <https://doi.org/10.2166/wqrj.2005.035>
- McMaster, M.E., Mark Hewitt, L., Parrott, J.L., 2006. A decade of research on the environmental impacts of pulp and paper mill effluents in Canada: field studies and mechanistic research. *J. Toxicol. Environ. Health Part B* 9, 319–339. <https://doi.org/10.1080/15287390500195869>
- Miller, M.B., Clough, A.M., Batson, J.N., Vachet, R.W., 2006. Transition metal binding to cod otolith proteins. *J. Exp. Mar. Biol. Ecol.* 329, 135–143. <https://doi.org/10.1016/j.jembe.2005.08.016>
- Mintenbeck, K., Brey, T., Jacob, U., Knust, R., Struck, U., 2008. How to account for the lipid effect on carbon stable-isotope ratio ($\delta^{13}\text{C}$): sample treatment effects and model bias. *J. Fish Biol.* 72, 815–830. <https://doi.org/10.1111/j.1095-8649.2007.01754.x>
- Morrison, D.F., 1990. Multivariate statistical methods, in: *Multivariate Statistical Methods*. pp. 495–495. <http://dx.doi.org/10.2307/2344132>
- Morrissey, C.A., Boldt, A., Mapstone, A., Newton, J., Ormerod, S.J., 2013. Stable isotopes as indicators of wastewater effects on the macroinvertebrates of urban rivers. *Hydrobiologia* 700, 231–244. <https://doi.org/10.1007/s10750-012-1233-7>
- Moslemi-Aqdam, M., Baker, L.F., Baltzer, J.L., Branfireun, B.A., Evans, M.S., Laird, B.D., Low, G., Low, M., Swanson, H.K., 2022. Understanding among-lake variability of mercury concentrations in Northern pike (*Esox lucius*): A whole-ecosystem study in subarctic lakes. *Sci. Total Environ.* 822, 153430. <https://doi.org/10.1016/j.scitotenv.2022.153430>
- Moslemi-Aqdam, M., Low, G., Low, M., Branfireun, B.A., Swanson, H.K., 2021. Catchments affect growth rate of Northern pike, *Esox lucius*, in subarctic lakes. *Aquat. Sci.* 83, 59. <https://doi.org/10.1007/s00027-021-00817-4>
- Moslemi-Aqdam, M., Low, G., Low, M., Laird, B.D., Branfireun, B.A., Swanson, H.K., 2023. Estimates, spatial variability, and environmental drivers of mercury biomagnification rates through lake food webs in the Canadian subarctic. *Environ. Res.* 217, 114835. <https://doi.org/10.1016/j.envres.2022.114835>
- Mugiya, Y., Hakomori, T., Hatsutori, K., 1991. Trace metal incorporation into otoliths and scales in the goldfish, *Carassius auratus*. *Comp. Biochem. Physiol. Part C Comp. Pharmacol.* 99, 327–331. [https://doi.org/10.1016/0742-8413\(91\)90250-W](https://doi.org/10.1016/0742-8413(91)90250-W)
- Muir, D., Bossi, R., Carlsson, P., Evans, M., De Silva, A., Halsall, C., Rauert, C., Herzke, D., Hung, H., Letcher, R., Rig  t, F., Roos, A., 2019. Levels and trends of poly- and perfluoroalkyl substances in the Arctic environment – an update. *Emerg. Contam.* 5, 240–271. <https://doi.org/10.1016/j.emcon.2019.06.002>
- Munkittrick, K.R., 1992. A review and evaluation of study design considerations for site-specifically assessing the health of fish populations. *J. Aquat. Ecosyst. Health* 1, 283–293. <https://doi.org/10.1007/BF00044170>

- Munkittrick, K.R., Arens, C.J., Lowell, R.B., Kaminski, G.P., 2009. A review of potential methods of determining critical effect size for designing environmental monitoring programs. *Environ. Toxicol. Chem.* 28, 1361–1371. <https://doi.org/10.1897/08-376.1>
- Munkittrick, K.R., Barrett, T.J., McMaster, M.E., 2010. Guidance for site-specifically assessing the health of fish populations with emphasis on Canada's environmental effects monitoring program. *Water Qual. Res. J.* 45, 209–221. <https://doi.org/10.2166/wqrj.2010.024>
- Munkittrick, K.R., McGeachy, S.A., McMaster, M.E., Courtenay, S.C., 2002. Overview of freshwater fish studies from the pulp and paper environmental effects monitoring program. *Water Qual. Res. J.* 37, 49–77. <https://doi.org/10.2166/wqrj.2002.005>
- Newell, S.L., Doubleday, N.C., Community of Chesterfield Inlet, Nunavut, 2020. Sharing country food: connecting health, food security and cultural continuity in Chesterfield Inlet, Nunavut. *Polar Res.* 39. <https://doi.org/10.33265/polar.v39.3755>
- Norstrom, R.J., McKinnon, A.E., deFreitas, A.S.W., 1976. A bioenergetics-based model for pollutant accumulation by fish. Simulation of PCB and methylmercury residue levels in Ottawa River Yellow perch (*Perca flavescens*). *J. Fish. Res. Board Can.* 33, 248–267. <https://doi.org/10.1139/f76-035>
- Nunami Stantec, 2011. Operations and maintenance manual for the water, sewage, and solid waste facilities, Baker Lake, NU (Licence No. 3BM-BAK1526), Nunavut Water Board. Rankin Inlet, Nunavut.
- Oksanen, J., Blanchet, F.G., Friendly, M., Kindt, R., Legendre, P., McGlinn, D., Minchin, P.R., O'hara, R.B., Simpson, G.L., Solymos, P., 2019. Community ecology package. R Package Version 2, 1–296. <https://doi.org/10.32614/CRAN.package.vegan>
- Oksanen, J., Kindt, R., Legendre, P., O'Hara, B., Stevens, M.H.H., Oksanen, M.J., Suggests, M., 2007. The vegan package. *Community Ecol. Package* 10, 719.
- Pannella, G., 1971. Fish Otoliths: Daily Growth Layers and Periodical Patterns. *Science* 173, 1124–1127. <https://doi.org/10.1126/science.173.4002.1124>
- Papadopoulou, C., Kanas, G.D., Kassimati, E.M., 1978. Zinc content in otoliths of mackerel from the Aegean. *Mar. Pollut. Bull.* 9, 106–108. [https://doi.org/10.1016/0025-326X\(78\)90482-4](https://doi.org/10.1016/0025-326X(78)90482-4)
- Paranjape, A.R., Hall, B.D., 2017. Recent advances in the study of mercury methylation in aquatic systems. *FACETS* 2, 85–119. <https://doi.org/10.1139/facets-2016-0027>
- Parks, J.W., Craig, P.J., Neary, B.P., Ozburn, G., Romani, D., 1991. Biomonitoring in the mercury-contaminated Wabigoon–English–Winnipeg River (Canada) system: selecting the best available bioindicator. *Appl. Organomet. Chem.* 5, 487–495. <https://doi.org/10.1002/aoc.590050606>
- Paton, C., Hellstrom, J., Paul, B., Woodhead, J., Hergt, J., 2011. Lolite: Freeware for the visualisation and processing of mass spectrometric data. *J. Anal. At. Spectrom.* 26, 2508–2518. <https://doi.org/10.1039/C1JA10172B>
- Payan, P., De Pontual, H., Bœuf, G., Mayer-Gostan, N., 2004. Endolymph chemistry and otolith growth in fish. *Comptes Rendus Palevol, BIOMINERALISATION : DIVERSITE ET UNITE* 3, 535–547. <https://doi.org/10.1016/j.crpv.2004.07.013>
- Peterson, B.J., Fry, B., 1987. Stable Isotopes in Ecosystem Studies. *Annu. Rev. Ecol. Syst.* 18, 293–320. <https://doi.org/10.1146/annurev.es.18.110187.001453>

- Pickhardt, P.C., Folt, C.L., Chen, C.Y., Klaue, B., Blum, J.D., 2002. Algal blooms reduce the uptake of toxic methylmercury in freshwater food webs. *Proc. Natl. Acad. Sci.* 99, 4419–4423. <https://doi.org/10.1073/pnas.072531099>
- Playle, R.C., Gensemer, R.W., Dixon, D.G., 1992. Copper accumulation on gills of fathead minnows: Influence of water hardness, complexation and pH of the gill micro-environment. *Environ. Toxicol. Chem.* 11, 381–391. <https://doi.org/10.1002/etc.5620110312>
- Pollock, M.S., Clarke, L.M.J., Dubé, M.G., 2007. The effects of hypoxia on fishes: from ecological relevance to physiological effects. *Environ. Rev.* 15, 1–14. <https://doi.org/10.1139/a06-006>
- Post, D.M., Layman, C.A., Arrington, D.A., Takimoto, G., Quattrochi, J., Montaña, C.G., 2007. Getting to the fat of the matter: models, methods and assumptions for dealing with lipids in stable isotope analyses. *Oecologia* 152, 179–189. <https://doi.org/10.1007/s00442-006-0630-x>
- Power, M., Klein, G.M., Guiguer, K.R.R.A., Kwan, M.K.H., 2002. Mercury accumulation in the fish community of a sub-Arctic lake in relation to trophic position and carbon sources. *J. Appl. Ecol.* 39, 819–830. <https://doi.org/10.1046/j.1365-2664.2002.00758.x>
- Proctor, C.H., Thresher, R.E., 1998. Effects of specimen handling and otolith preparation on concentration of elements in fish otoliths. *Mar. Biol.* 131, 681–694. <https://doi.org/10.1007/s002270050360>
- Radtke, R., Svenning, M., Malone, D., Klements, A., Ruzicka, J., Fey, D., 1996. Migrations in an extreme northern population of Arctic charr *Salvelinus alpinus*: insights from otolith microchemistry. *Mar. Ecol. Prog. Ser.* 136, 13–23. <http://dx.doi.org/10.3354/meps136013>
- Radtke, R.L., Shafer, D.J., 1992. Environmental sensitivity of fish otolith microchemistry. *Mar. Freshw. Res.* 43, 935–951. <https://doi.org/10.1071/MF9920935>
- Ragush, C.M., Schmidt, J.J., Krkosek, W.H., Gagnon, G.A., Truelstrup-Hansen, L., Jamieson, R.C., 2015. Performance of municipal waste stabilization ponds in the Canadian Arctic. *Ecol. Eng.* 83, 413–421. <https://doi.org/10.1016/j.ecoleng.2015.07.008>
- Ranaldi, M.M., Gagnon, M.M., 2008a. Trace metal incorporation in otoliths of Black bream (*Acanthopagrus butcheri* Munro), an indicator of exposure to metal contamination. *Water. Air. Soil Pollut.* 194, 31–43. <https://doi.org/10.1007/s11270-008-9696-x>
- Ranaldi, M.M., Gagnon, M.M., 2008b. Zinc incorporation in the otoliths of juvenile Pink snapper (*Pagrus auratus* Forster): the influence of dietary versus waterborne sources. *J. Exp. Mar. Biol. Ecol.* 360, 56–62. <https://doi.org/10.1016/j.jembe.2008.03.013>
- Reed, R.J., 1964. Life history and migration patterns of Arctic grayling, *Thymallus arcticus*, (Pallas), in the Tanana River drainage of Alaska. Alaska. Department of Fish and Game. [https://www.adfg.alaska.gov/fedaidpdfs/fredf-9-12\(21\)R-I.pdf](https://www.adfg.alaska.gov/fedaidpdfs/fredf-9-12(21)R-I.pdf)
- Reis-Santos, P., Gillanders, B.M., Sturrock, A.M., Izzo, C., Oxman, D.S., Lueders-Dumont, J.A., Hüsey, K., Tanner, S.E., Rogers, T., Doubleday, Z.A., Andrews, A.H., Trueman, C., Brophy, D., Thiem, J.D., Baumgartner, L.J., Willmes, M., Chung, M.-T., Charapata, P., Johnson, R.C., Trumble, S., Heimbrand, Y., Limburg, K.E., Walther, B.D., 2023. Reading the biomineralized book of life: expanding otolith biogeochemical research and applications

- for fisheries and ecosystem-based management. *Rev. Fish Biol. Fish.* 33, 411–449.
<https://doi.org/10.1007/s11160-022-09720-z>
- Reis-Santos, P., Vasconcelos, R.P., Tanner, S.E., Fonseca, V.F., Cabral, H.N., Gillanders, B.M., 2018. Extrinsic and intrinsic factors shape the ability of using otolith chemistry to characterize estuarine environmental histories. *Mar. Environ. Res.* 140, 332–341.
<https://doi.org/10.1016/j.marenvres.2018.06.002>
- Reynolds, J.B., Simmons, R.C., Burkholder, A.R., 1989. Effects of placer mining discharge on health and food of Arctic grayling. *JAWRA J. Am. Water Resour. Assoc.* 25, 625–635.
<https://doi.org/10.1111/j.1752-1688.1989.tb03100.x>
- Ribey, S.C., Munkittrick, K.R., McMaster, M.E., Courtenay, S., Langlois, C., Munger, S., Rosaasen, A., Whitley, G., 2002. Development of a monitoring design for examining effects in wild fish associated with discharges from metal mines. *Water Qual. Res. J.* 37, 229–249.
<https://doi.org/10.2166/wqrj.2002.015>
- Ricker, W.E., 1975. Computation and interpretation of biological statistics of fish populations. *Fish Res. Board Can. Bull.* 191, 1–382. <https://publications.gc.ca/pub?id=9.581563&sl=0>
- Robin, T., Burnett, K., Parker, B., Skinner, K., 2021. Safe food, dangerous Lands? Traditional foods and Indigenous Peoples in Canada. *Front. Commun.* 6.
<https://doi.org/10.3389/fcomm.2021.749944>
- Robinson, C.S., Tetreault, G.R., McMaster, M.E., Servos, M.R., 2016. Impacts of a tertiary treated municipal wastewater effluent on the carbon and nitrogen stable isotope signatures of two darter species (*Etheostoma blennioides* and *E. caeruleum*) in a small receiving environment. *Ecol. Indic.* 60, 594–602.
<https://doi.org/10.1016/j.ecolind.2015.06.041>
- Ruttenberg, B., Hamilton, S., Hickford, M., Paradis, G., Sheehy, M., Standish, J., Ben-Tzvi, O., Warner, R., 2005. Elevated levels of trace elements in cores of otoliths and their potential for use as natural tags. *Mar. Ecol. Prog. Ser.* 297, 273–281.
<https://doi.org/10.3354/meps297273>
- Santore, R.C., Di Toro, D.M., Paquin, P.R., Allen, H.E., Meyer, J.S., 2001. Biotic ligand model of the acute toxicity of metals. 2. Application to acute copper toxicity in freshwater fish and *Daphnia*. *Environ. Toxicol. Chem.* 20, 2397–2402.
<https://doi.org/10.1002/etc.5620201035>
- Schaefer, C.M., Deslauriers, D., Jeffries, K.M., 2022. The truncate soft-shell clam, *Mya truncata*, as a biomonitor of municipal wastewater exposure and historical anthropogenic impacts in the Canadian Arctic. *Can. J. Fish. Aquat. Sci.* 79, 367–379.
<https://doi.org/10.1139/cjfas-2021-0078>
- Schindler, D.W., Kidd, K.A., Muir, D.C.G., Lockhart, W.L., 1995. The effects of ecosystem characteristics on contaminant distribution in northern freshwater lakes. *Sci. Total Environ., Ecological Effects of Arctic Airborne Contaminants* 160–161, 1–17.
[https://doi.org/10.1016/0048-9697\(95\)04340-7](https://doi.org/10.1016/0048-9697(95)04340-7)
- Schmidt, J.J., Ragush, C.M., Krkosek, W.H., Gagnon, G.A., Jamieson, R.C., 2016. Characterizing phosphorus removal in passive waste stabilization ponds in Arctic communities. *Arct. Sci.* 2, 1–14. <https://doi.org/10.1139/as-2015-0002>
- Scott, W.B., Crossman, E.J., 1973. Freshwater fishes of Canada. *Fish. Res. Board Can.* 184, 1–966. <https://publications.gc.ca/pub?id=9.870340&sl=0>

- Simoneau, M., Lucotte, M., Garceau, S., Laliberté, D., 2005. Fish growth rates modulate mercury concentrations in walleye (*Sander vitreus*) from eastern Canadian lakes. *Environ. Res.* 98, 73–82. <https://doi.org/10.1016/j.envres.2004.08.002>
- Skei, J., Larsson, P., Rosenberg, R., Jonsson, P., Olsson, M., Broman, D., 2000. Eutrophication and contaminants in aquatic ecosystems. *AMBIO J. Hum. Environ.* 29, 184–194. <https://doi.org/10.1579/0044-7447-29.4.184>
- Slack, R.J., Gronow, J.R., Voulvoulis, N., 2005. Household hazardous waste in municipal landfills: contaminants in leachate. *Sci. Total Environ.* 337, 119–137. <https://doi.org/10.1016/j.scitotenv.2004.07.002>
- Stewart, D.B., Mochnacz, N.J., Reist, J.D., Carmichael, T.J., Sawatzky, C.D., 2007a. Fish life history and habitat use in the Northwest Territories: Arctic grayling (*Thymallus arcticus*). *Can. Manuscri. Rep. Fish. Aquat. Sci.* 2797, 64. <https://publications.gc.ca/pub?id=9.618785&sl=0>
- Stewart, D.B., Mochnacz, N.J., Reist, J.D., Carmichael, T.J., Sawatzky, C.D., 2007b. Fish diets and food webs in the Northwest Territories: Arctic grayling (*Thymallus arcticus*). *Can. Manuscri. Rep. Fish. Aquat. Sci.* 2796, 21. <https://publications.gc.ca/pub?id=9.618784&sl=0>
- Stroski, K.M., Luong, K.H., Challis, J.K., Chaves-Barquero, L.G., Hanson, M.L., Wong, C.S., 2020. Wastewater sources of per- and polyfluorinated alkyl substances (PFAS) and pharmaceuticals in four Canadian Arctic communities. *Sci. Total Environ.* 708, 134494. <https://doi.org/10.1016/j.scitotenv.2019.134494>
- Sturrock, A., Trueman, C., Milton, J., Waring, C., Cooper, M., Hunter, E., 2014. Physiological influences can outweigh environmental signals in otolith microchemistry research. *Mar. Ecol. Prog. Ser.* 500, 245–264. <https://doi.org/10.3354/meps10699>
- Sturrock, A.M., Hunter, E., Milton, J.A., Eimf, Johnson, R.C., Waring, C.P., Trueman, C.N., 2015. Quantifying physiological influences on otolith microchemistry. *Methods Ecol. Evol.* 6, 806–816. <https://doi.org/10.1111/2041-210X.12381>
- Sunda, W.G., Lewis, J.A.M., 1978. Effect of complexation by natural organic ligands on the toxicity of copper to a unicellular Alga, *Monochrysis lutheri*. *Limnol. Oceanogr.* 23, 870–876. <https://doi.org/10.4319/lo.1978.23.5.0870>
- Swanson, H.K., Kidd, K.A., Babaluk, J.A., Wastle, R.J., Yang, P.P., Halden, N.M., Reist, J.D., 2010. Anadromy in Arctic populations of lake trout (*Salvelinus namaycush*) : otolith microchemistry, stable isotopes, and comparisons with Arctic char (*Salvelinus alpinus*). *Can. J. Fish. Aquat. Sci.* 67, 842–853. <https://doi.org/10.1139/F10-022>
- Tessier, A., Campbell, P.G.C., 1987. Partitioning of trace metals in sediments: relationships with bioavailability. *Hydrobiologia* 149, 43–52. <https://doi.org/10.1007/BF00048645>
- Tetreault, G.R., Bennett, C.J., Shires, K., Knight, B., Servos, M.R., McMaster, M.E., 2011. Intersex and reproductive impairment of wild fish exposed to multiple municipal wastewater discharges. *Aquat. Toxicol.* 104, 278–290. <https://doi.org/10.1016/j.aquatox.2011.05.008>
- Tetreault, G.R., Brown, C.J., Bennett, C.J., Oakes, K.D., McMaster, M.E., Servos, M.R., 2013. Fish community responses to multiple municipal wastewater inputs in a watershed. *Integr. Environ. Assess. Manag.* 9, 456–468. <https://doi.org/10.1002/ieam.1364>

- Thomas, O.R.B., Ganio, K., Roberts, B.R., Swearer, S.E., 2017. Trace element–protein interactions in endolymph from the inner ear of fish: implications for environmental reconstructions using fish otolith chemistry. *Metallomics* 9, 239–249. <https://doi.org/10.1039/C6MT00189K>
- Thomas, O.R.B., Swearer, S.E., 2019. Otolith biochemistry—a review. *Rev. Fish. Sci. Aquac.* 27, 458–489. <https://doi.org/10.1080/23308249.2019.1627285>
- Thomas, O.R.B., Swearer, S.E., Kapp, E.A., Peng, P., Tonkin-Hill, G.Q., Papenfuss, A., Roberts, A., Bernard, P., Roberts, B.R., 2019. The inner ear proteome of fish. *FEBS J.* 286, 66–81. <https://doi.org/10.1111/febs.14715>
- Thomas, O.R.B., Thomas, K.V., Jenkins, G.P., Swearer, S.E., 2020. Spatio-temporal resolution of spawning and larval nursery habitats using otolith microchemistry is element dependent. *Mar. Ecol. Prog. Ser.* 636, 169–187. <https://doi.org/10.3354/meps13229>
- Thomas, S.M., Kiljunen, M., Malinen, T., Eloranta, A.P., Amundsen, P.-A., Lodenius, M., Kahilainen, K.K., 2016. Food-web structure and mercury dynamics in a large subarctic lake following multiple species introductions. *Freshw. Biol.* 61, 500–517. <https://doi.org/10.1111/fwb.12723>
- Thomas, S.M., Melles, S.J., Mackereth, R.W., Tunney, T.D., Chu, C., Oswald, C.J., Bhavsar, S.P., Johnston, T.A., 2020. Climate and landscape conditions indirectly affect fish mercury levels by altering lake water chemistry and fish size. *Environ. Res.* 188, 109750. <https://doi.org/10.1016/j.envres.2020.109750>
- Thorrold, S.R., Swearer, S.E., 2009. Otolith chemistry, in: Green, B.S., Mapstone, B.D., Carlos, G., Begg, G.A. (Eds.), *Tropical Fish Otoliths: Information for Assessment, Management and Ecology*. Springer Netherlands, Dordrecht, pp. 249–295. https://doi.org/10.1007/978-1-4020-5775-5_8
- Thresher, R.E., 1999. Elemental composition of otoliths as a stock delineator in fishes. *Fish. Res.* 43, 165–204. [https://doi.org/10.1016/S0165-7836\(99\)00072-7](https://doi.org/10.1016/S0165-7836(99)00072-7)
- Tohse, H., Murayama, E., Ohira, T., Takagi, Y., Nagasawa, H., 2006. Localization and diurnal variations of carbonic anhydrase mRNA expression in the inner ear of the rainbow trout *Oncorhynchus mykiss*. *Comp. Biochem. Physiol. B Biochem. Mol. Biol.* 145, 257–264. <https://doi.org/10.1016/j.cbpb.2006.06.011>
- Trudel, M., Rasmussen, J.B., 2006. Bioenergetics and mercury dynamics in fish: a modelling perspective. *Can. J. Fish. Aquat. Sci.* 63, 1890–1902. <https://doi.org/10.1139/f06-081>
- Trudel, M., Rasmussen, J.B., 1997. Modeling the elimination of mercury by fish. *Environ. Sci. Technol.* 31, 1716–1722. <https://doi.org/10.1021/es960609t>
- U.S. EPA, 2007. EPA Method 7473 (SW-846): Mercury in Solids and Solutions by Thermal Decomposition, Amalgamation, and Atomic Absorption Spectrophotometry.
- U.S. EPA, 1998. Method 1630: Methyl Mercury in Water by Distillation, Aqueous Ethylation, Purge and Trap, and Cold Vapor Atomic Fluorescence Spectrometry.
- U.S. EPA, 1996a. Method 1669: Sampling Ambient Water for Trace Metals at EPA Water Quality Criteria Levels.
- U.S. EPA, 1996b. SW-846 Test Method 3052: Microwave Assisted Acid Digestion of Siliceous and Organically Based Matrices.

- U.S. EPA, 1994. Method 200.8: Determination of Trace Elements in Waters and Wastes by Inductively Coupled Plasma-Mass Spectrometry. <https://doi.org/10.1016/B978-0-8155-1398-8.50011-2>
- Vadeboncoeur, Y., Lodge, D.M., Carpenter, S.R., 2001. Whole-lake fertilization effects on distribution of primary production between benthic and pelagic habitats. *Ecology* 82, 1065–1077. [https://doi.org/10.1890/0012-9658\(2001\)082\[1065:WLFEOD\]2.0.CO;2](https://doi.org/10.1890/0012-9658(2001)082[1065:WLFEOD]2.0.CO;2)
- van Achterbergh, E., Ryan, C.G., Griffin, W.L., 1999. GLITTER: On-line Interactive Data Reduction for the Laser Ablation Inductively Coupled Plasma Mass Spectrometry Microprobe. Presented at the Ninth Annual V. M. Goldschmidt Conference. <https://ui.adsabs.harvard.edu/abs/1999nag.conf.7215V>
- Vignon, M., Tabouret, H., Aymes, J.-C., Pecheyran, C., Rives, J., Coste-Heinrich, P., Huchet, E., Bareille, G., 2023. Effect of metabolic rate on time-lag changes in otolith microchemistry: an experimental approach using *Salmo trutta*. *J. Exp. Biol.* 226, jeb245265. <https://doi.org/10.1242/jeb.245265>
- Vymazal, J., 2007. Removal of nutrients in various types of constructed wetlands. *Sci. Total Environ., Contaminants in Natural and Constructed Wetlands: Pollutant Dynamics and Control* 380, 48–65. <https://doi.org/10.1016/j.scitotenv.2006.09.014>
- Vymazal, J., Krása, P., 2003. Distribution of Mn, Al, Cu and Zn in a constructed wetland receiving municipal sewage. *Water Sci. Technol.* 48, 299–305. <http://dx.doi.org/10.2166/wst.2003.0336>
- Wang, R., Wang, W.-X., 2012. Contrasting mercury accumulation patterns in tilapia (*Oreochromis niloticus*) and implications on somatic growth dilution. *Aquat. Toxicol.* 114–115, 23–30. <https://doi.org/10.1016/j.aquatox.2012.02.014>
- Watras, C.J., Back, R.C., Halvorsen, S., Hudson, R.J.M., Morrison, K.A., Wentz, S.P., 1998. Bioaccumulation of mercury in pelagic freshwater food webs. *Sci. Total Environ.* 219, 183–208. [https://doi.org/10.1016/S0048-9697\(98\)00228-9](https://doi.org/10.1016/S0048-9697(98)00228-9)
- Wayland, M., Hobson, K., 2001. Stable carbon, nitrogen, and sulfur isotope ratios in riparian food webs on rivers receiving sewage and pulp-mill effluents. *Can. J. Zool.-Rev. Can. Zool. - CAN J ZOOL* 79, 5–15. <https://doi.org/10.1139/cjz-79-1-5>
- Weber, K.A., Urrutia, M.M., Churchill, P.F., Kukkadapu, R.K., Roden, E.E., 2006. Anaerobic redox cycling of iron by freshwater sediment microorganisms. *Environ. Microbiol.* 8, 100–113. <https://doi.org/10.1111/j.1462-2920.2005.00873.x>
- Weigele, J., Franz-Odenaal, T.A., Hilbig, R., 2016. Not all inner ears are the same: otolith matrix proteins in the inner ear of sub-adult cichlid fish, *Oreochromis Mossambicus*, reveal insights into the biomineralization process. *Anat. Rec.* 299, 234–245. <https://doi.org/10.1002/ar.23289>
- Westerlund, C., 2007. Road runoff quality in cold climates (PhD Thesis). Luleå tekniska universitet.
- Wetzel, R.G., 2001. Limnology: lake and river ecosystems. Gulf Professional Publishing.
- Wood, C.M., Farrell, A.P., Brauner, C.J., 2012. Homeostasis and toxicology of essential metals. Academic Press.
- Wu, S., Vymazal, J., Brix, H., 2019. Critical review: biogeochemical networking of iron in constructed wetlands for wastewater treatment. *Environ. Sci. Technol.* 53, 7930–7944. <https://doi.org/10.1021/acs.est.9b00958>

- Xue, H., Kistler, D., Sigg, L., 1995. Competition of copper and zinc for strong ligands in a eutrophic lake. *Limnol. Oceanogr.* 40, 1142–1152.
<https://doi.org/10.4319/lo.1995.40.6.1142>
- Yates, C.N., 2012. Developing an understanding for wastewater treatment in remote communities in Nunavut, Canada: investigating the performance, planning practice and function of tundra and constructed treatment wetlands (Doctoral Thesis). University of Waterloo.
- Yates, C.N., Wootton, B., Murphy, S.D., 2020. Wastewater treatment wetlands: use in Arctic regions 5-year update, in: *Managing Water Resources and Hydrological Systems*. CRC Press. <http://dx.doi.org/10.1201/9781003045045-60>
- Yates, C.N., Wootton, B.C., Murphy, S.D., 2012. Performance assessment of arctic tundra municipal wastewater treatment wetlands through an arctic summer. *Ecol. Eng.* 44, 160–173. <https://doi.org/10.1016/j.ecoleng.2012.04.011>
- Zhu, L., Anello, R., Rühland, K.M., Pisaric, M.F.J., Kokelj, S.V., Prince, T., Smol, J.P., 2019. Impacts of Road Dust on Small Subarctic Lake Systems. *Arctic* 72, 434–457.
<https://doi.org/10.14430/arctic69527>
- Zimmerman, C.E., 2005. Relationship of otolith strontium-to-calcium ratios and salinity: experimental validation for juvenile salmonids. *Can. J. Fish. Aquat. Sci.* 62, 88–97.
<https://doi.org/10.1139/f04-182>
- Zimmerman, C.E., Reeves, G.H., 2000. Population structure of sympatric anadromous and nonanadromous *Oncorhynchus mykiss* : evidence from spawning surveys and otolith microchemistry. *Can. J. Fish. Aquat. Sci.* 57, 2152–2162. <https://doi.org/10.1139/f00-192>
- Zuur, A.F., Ieno, E.N., Walker, N., Saveliev, A.A., Smith, G.M., 2009. *Mixed effects models and extensions in ecology with R*, Statistics for Biology and Health. Springer, New York, NY.
<https://doi.org/10.1007/978-0-387-87458-6>

Appendices

Supplementary information for Chapter 1

Table S1.1 Historical water quality conditions in Airplane Lake, Baker Lake, Nunavut: the third and final lake within the lagoon-tundra wastewater wetland treatment system. Data sourced from the Nunavut Impact Review Board (NIRB) public registry.

Year	Month	Day	Analysis	Site	mg/L										
					Total P	NO ₃ ⁻ + NO ₂ ⁻	NH ₃	Al	Cu	Fe	Ba	Mn	Pb	Se	Zn
1994	6	8	INAC* Analytical Laboratory, Yellowknife	Airplane inflow	Na	0.055	0.055	Na	0.004	0.577	Na	0.0649	0.001	Na	0.006
2006	9	Na	Nunami Jacques Whitford Ltd.	Bak-3**	0.019	0.448	0.024	0.011	0.002	0.09	0.035	0.007	<0.001	0.001	<0.010
2007	<u>7</u>	Na	Nunami Jacques Whitford Ltd.	Bak-3	0.131	0.097	0.911	0.121	0.002	0.12	0.015	0.030	<0.001	<0.001	<0.001
2010	10	Na	Nunami Jacques Whitford Ltd.	Bak-3	0.047	0.071	0.050	0.030	0.002	0.002	0.051	0.011	<0.001	<0.001	<0.005
2015	<u>6</u>	29	ALS Environmental, Winnipeg	Bak-3	0.145	0.079	0.247	0.047	0.002	0.47	Na	0.060	<0.001	Na	0.002
2017	<u>6</u>	15	ALS Environmental, Winnipeg	Bak-3	0.090	0.070	0.202	0.063	0.001	0.426	Na	0.038	<0.001	Na	0.002
2018	<u>6</u>	21	ALS Environmental, Winnipeg	Bak-3	0.176	0.070	1.190	Na	0.003	0.383	Na	0.069	<0.001	Na	0.004
2019	8	26	BASL, UOA; Biotron, UWO	Air-WQ01 [#]	0.092	0.090	Na	0.2213	0.018	0.016	0.120	0.002	<0.001	<0.001	0.040
2021			BASL, UOA; Biotron, UWO	Air-WQ01 [#]											

Bold implies a Canadian Environmental Quality (CCME) guidance exceedance.

Na refers to unavailable data.

* Indian and Northern Affairs Canada (now dissolved)

** Airplane Lake outflow to Baker Lake Creek

South-West side of Airplane Lake, towards Baker Lake Creek inflow

Supplementary information for Chapter 2

Table S2.1 Literature review of metals chosen for analysis, emphasizing those detectable in tissues (i.e., liver, other tissues, otolith) and commonly found in landfill or municipal wastewater. **LF** – Landfill; **WW** – Wastewater, X – depicts if the tissue will be analyzed for the purpose of this research project.

Metal	Liver	Tissue	Otolith	Potential Source*	Water chemistry factors affecting bioavailability	Comments/Justification
Cadmium (Cd)	X		X	LF, WW ¹	Salinity, DOC, [H ⁺], [Ca ²⁺] ²	Enters fish via the gills and accumulates most heavily in detoxifying organs (kidney, liver) ^{3, 20} .
Copper (Cu)	X		X	LF ⁴ , WW ⁵	Temperature, DOC, alkalinity, pH, hardness ⁶	Essential for metabolic function but can be toxic if accumulated more than cellular needs. The liver is the major organ involved in Cu homeostasis leading to elevated concentrations in the liver compared to other tissues ^{7, 20} .
Nickel (Ni)	X		X	LF ⁴ , WW ⁵	pH, alkalinity, hardness ⁶	Highest concentrations of Ni are observed in fish livers which is a major producer of metal binding proteins ⁸ .
Lead (Pb)	X		X	LF ⁴ , WW ⁵	pH, alkalinity, hardness ⁶	Deposits in various organs with high levels found in livers and bones ^{8, 20} .
Zinc (Zn)	X		X	LF ⁴ , WW ⁵	Alkalinity, hardness ⁶	Shows affinity to gonads because of its metabolic role in this tissue but also concentrates in the liver ^{8, 20} .
Mercury (Hg)	X	X	X	Some evidence for LF ⁹	[Al], DOC, pH ¹⁰	Predominantly concentrated in detoxifying organs (liver, kidney); organic methylmercury binds to proteins and is distributed throughout different organs ^{3, 20} . It is important to analyze mercury in edible tissue because of its toxic effects at [high].
Selenium (Se)	X	X	X	LF ¹¹	DOC, pH ¹²	Strongly bioconcentrated in the kidney and liver of fish ³ . However, Se has important nutritional benefits, and concentrations should be quantified in edible fish tissue ¹³ .
Strontium (Sr)		X	X	LF ¹⁴	Temperature, salinity ¹²	Sr is considered a non-essential element to fish since its metabolic role is unclear. Wild fish muscle has been reported to have elevated [Sr] over liver ¹⁵ . Otolith Sr profiles have been used to investigate movement of anadromous fish ¹⁶ .
Iron (Fe)	X		X	WW ⁵	Alkalinity, DOC, pH ¹⁵	Essential element for cellular respiration and oxygen transfer but can be toxic at [high]. Fe homeostasis occurs in the liver where [high] are observed in fish ¹⁵ .
Arsenic (As)	X		X	WW ⁵	Temperature ¹²	As is mostly concentrated in the liver and kidneys, which is expected since metalloids are actively eliminated by fish ³ .
Aluminum (Al)	X		X	LF ¹⁷	pH ¹⁸	Adsorption of Al the gill surface is rapid and the primary cause of acute and toxic effects. Uptake of Al from the water to internal organs is slow but has been documented at [high] ¹⁸
Manganese (Mn)	X		X	WW ⁵	DOC ¹²	Mn is an essential element stored predominantly in fish skin and gills. Concentrations of Mn in liver is higher than in muscle for multiple fish species ¹⁹ .

Table S2.1 References.

1. Choi SB and Yun Y. 2006. Biosorption of cadmium by various types of dried sludge: An equilibrium study and investigation of mechanisms. *J Hazard Mater* 138(2):378-83.
2. Wright D and Welbourn P. 1994. Cadmium in the aquatic environment: A review of ecological, physiological, and toxicological effects on biota. *Env Rev* 2(2):187-214.
3. Mason R, Laporte J, Andres S. 2000. Factors controlling the bioaccumulation of mercury, methylmercury, arsenic, selenium, and cadmium by freshwater invertebrates and fish. *Arch Environ Contam Toxicol* 38(3):283-97.
4. Christensen JB, Jensen DL, Christensen TH. 1996. Effect of dissolved organic carbon on the mobility of cadmium, nickel and zinc in leachate polluted groundwater. *Water Res* 30(12):3037-49.
5. da Silva Oliveira A, Bocio A, Trevilato TMB, Takayanagui AMM, Domingo JL, Segura-Muñoz SI. 2007. Heavy metals in untreated/treated urban effluent and sludge from a biological wastewater treatment plant. *Environ Sci and Pol* 14(7):483.
6. Meyer JS. 2007. Effects of water chemistry on bioavailability and toxicity of waterborne cadmium, copper, nickel, lead, and zinc to freshwater organisms. SETAC Press. 328 p.
7. Bury NR, Walker PA, Glover CN. 2003. Nutritive metal uptake in teleost fish. *J Exp Biol* 206(1):11-23.
8. Jezierska B and Witeska M. 2006. The metal uptake and accumulation in fish living in polluted waters. In: *Soil and water pollution monitoring, protection and remediation*. Springer. 107 p.
9. Slack R, Gronow J, Voulvoulis N. 2005. Household hazardous waste in municipal landfills: Contaminants in leachate. *Sci Total Environ* 337(1-3):119-37.
10. Kidd KA, Muir DC, Evans MS, Wang X, Whittle M, Swanson HK, Johnston T, Guildford S. 2012. Biomagnification of mercury through lake trout (*salvelinus namaycush*) food webs of lakes with different physical, chemical and biological characteristics. *Sci Total Environ* 438:135-43.
11. Lemly AD. 2004. Aquatic selenium pollution is a global environmental safety issue. *Ecotoxicol Environ Saf* 59(1):44-56.
12. Hoffman DJ, Rattner BA, Burton Jr GA, Cairns Jr J. 2002. *Handbook of ecotoxicology*. CRC press. 1315 p.
13. Reyes ES, Aristizabal Henao JJ, Kornobis KM, Hanning RM, Majowicz SE, Liber K, Stark KD, Low G, Swanson HK, Laird BD. 2017. Associations between omega-3 fatty acids, selenium content, and mercury levels in wild-harvested fish from the Dehcho region, Northwest Territories, Canada. *J of Tox and Environ Health* 80(1):18-31.
14. Nigro A, Sappa G, Barbieri M. 2017. Strontium isotope as tracers of groundwater contamination. *Procedia Earth Planet Sci* 17:352-5.
15. Fallah AA, Saei-Dehkordi SS, Nematollahi A, Jafari T. 2011. Comparative study of heavy metal and trace element accumulation in edible tissues of farmed and wild rainbow trout (*oncorhynchus mykiss*) using ICP-OES technique. *Microchem Journal* 98(2):275-9.
16. Swanson HK, Kidd KA, Babaluk JA, Wastle RJ, Yang PP, Halden NM, Reist JD. 2010. Anadromy in arctic populations of lake trout (*salvelinus namaycush*): Otolith microchemistry, stable isotopes, and comparisons with arctic char (*salvelinus alpinus*). *Can J Fish Aquat Sci* 67(5):842-53.
17. Stark TD, Martin JW, Gerbasi GT, Thalhamer T, Gortner RE. 2012. Aluminum waste reaction indicators in a municipal solid waste landfill. *J Geotech Geoenviron Eng* 138(3):252-61.
18. Wilson RW. 2011. Aluminum. In: *Fish physiology*. Elsevier. 67 p.
19. Rajkowska M and Protasowicki M. 2013. Distribution of metals (Fe, Mn, Zn, Cu) in fish tissues in two lakes of different trophy in northwestern Poland. *Environ Monit Assess* 185(4):3493-502.
20. Farkas A, Salanki J, Varanka I. 2000. Heavy metal concentrations in fish of Lake Balaton. *Lakes Reservoirs* 5(4):271-9.

Table S2.2 Sample quality assurance-quality control (QA/QC) was conducted in accordance with international standards. CRM – reference material recoveries, SR – spike recoveries, MDL – method detection limit, RPD – relative percent difference. Values are mean $\pm$ standard error.

Analysis	Parameter	Fish (muscle)	Fish (liver)	Bulk Seston	Water	Sediment
$\delta^{15}\text{N}$	AD	0.304 $\pm$ 0.047 ‰	-	-	-	-
	RPD	2.520 $\pm$ 0.447 %				
	n	11				
$\delta^{13}\text{C}$	AD	0.094 $\pm$ 0.025 ‰	-	-	-	-
	RPD	0.461 $\pm$ 0.123 %				
	n	11				
Hg	RPD	3.303 $\pm$ 0.882 %	4.987 $\pm$ 0.816 %	4.444 $\pm$ 0.408 %	4.000 $\pm$ 0.000 %	4.600 $\pm$ 0.577 %
	CRM	104.457 $\pm$ 1.155 %	103.519 $\pm$ 1.225 %	105.889 $\pm$ 0.816 %	105.000 $\pm$ 0.000 %	105.500 $\pm$ 1.453 %
	SR	99.096 $\pm$ 2.186 %	98.064 $\pm$ 4.082 %	100.333 $\pm$ 2.449 %	103.000 $\pm$ 0.000 %	99.600 $\pm$ 2.906 %
	n	188	156	10	23	20
	MDL	0.025 $\pm$ 0.002 %	10.701 $\pm$ 0.004 ng	0.067 $\pm$ 0.003 %	0.064 $\pm$ 0.000 ng/L	0.067 $\pm$ 0.003 %
MeHg	RPD	5.723 $\pm$ 1.00 %	10.455 $\pm$ 2.858 %	7.000 $\pm$ 0.000 %	7.000 $\pm$ 0.000 %	7.700 $\pm$ 2.333 %
	CRM	98.367 $\pm$ 2.186 %	93.115 $\pm$ 7.348 %	98.889 $\pm$ 2.858 %	102.000 $\pm$ 0.000 %	96.600 $\pm$ 5.196 %
	SR	111.761 $\pm$ 7.000 %	105.128 $\pm$ 8.165 %	105.667 $\pm$ 8.573 %	115.000 $\pm$ 0.000 %	104.600 $\pm$ 6.839 %
	n	188	156	10	23	20
	MDL	0.051 $\pm$ 0.015	1.007 $\pm$ 0.001 ng	0.006 $\pm$ 0.001 ng	0.007 $\pm$ 0.000 ng/L	0.006 $\pm$ 0.001 ng
ICP-MS	RPD	w/in 20% control limit	w/in 20% control limit	w/in 20% control limit	w/in 20% control limit	w/in 20% control limit
	CRM	w/in 15% expected value	w/in 15% expected value	w/in 15% expected value	w/in 15% expected value	w/in 15% expected value
	SR	w/in 30% known value	w/in 30% known value	w/in 30% known value	w/in 25% known value	w/in 30% known value
	n	159	156	10	46	20

Table S2.3 Summary of abiotic variables in six lakes of Baker Lake, Nunavut, Canada. Upper Lagoon Lake is situated upstream of the wastewater wetland, while Lagoon Lake, Finger Lake, and Airplane Lake constitute the lagoon-tundra wastewater wetland. Lake R1 and Lake R2 serve as reference systems. Values are mean $\pm$ standard error.

Parameter	Upper Lagoon	Lagoon	Finger	Airplane	R1	R2
Latitude (N)	64°20'20"N	64°19'53"N	64°19'44"N	64°19'11"N	64°20'26"N	64°19'32"N
Longitude (W)	96° 1'11"W	96° 0'12"W	95°59'28"W	95°57'45"W	95°55'8"W	96° 7'1"W
Surface Area (km ²)	0.16	0.02	0.05	0.61	0.12	0.26
Water						
Temperature (°C)	10.20 $\pm$ 0.03	14.63 $\pm$ 0.64	11.00 $\pm$ 0.07	8.60 $\pm$ 0.04	13.60 $\pm$ 0.05	9.90 $\pm$ 0.05
pH	6.600 $\pm$ 0.04	6.090 $\pm$ 0.01	6.448 $\pm$ 0.08	6.533 $\pm$ 0.10	6.993 $\pm$ 0.03	76.503 $\pm$ 0.03
Specific cond. (μ S/m)	63.650 $\pm$ 0.25	123.50 $\pm$ 3.04	83.90 $\pm$ 1.03	65.60 $\pm$ 0.58	50.50 $\pm$ 0.42	49.80 $\pm$ 0.49
DO (mg/L)	11.030 $\pm$ 0.10	3.85 $\pm$ 0.46	10.392 $\pm$ 0.22	12.217 $\pm$ 0.39	11.783 $\pm$ 0.12	11.083 $\pm$ 0.13
DOC	4.570 [†]	7.080 [†]	8.160 $\pm$ 3.81	8.880 $\pm$ 0.33	5.600 $\pm$ 0.26	5.740 $\pm$ 0.09
DIC	4.550 [†]	5.100 [†]	5.580 $\pm$ 0.11	2.630 $\pm$ 0.27	5.330 $\pm$ 2.27	3.840 $\pm$ 0.26
SO ₄ (mg/L)	1.690 $\pm$ 0.08	4.080 $\pm$ 0.65	16.248 $\pm$ 0.26	5.467 $\pm$ 0.14	2.165 $\pm$ 0.11	1.735 $\pm$ 0.09
Chlorine (mg/L)	4.880 $\pm$ 0.13	10.250 $\pm$ 0.65	9.530 $\pm$ 0.29	11.290 $\pm$ 0.21	4.245 $\pm$ 0.12	9.983 $\pm$ 0.31
Calcium (mg/L)	8.580 $\pm$ 0.16	7.410 [†]	11.290 $\pm$ 0.31	7.863 $\pm$ 0.07	6.490 $\pm$ 0.29	9.090 $\pm$ 0.55
TN (mg/L)	293.500 $\pm$ 12.50	5790.000 $\pm$ 629.33	2065.000 $\pm$ 110.94	1129.200 $\pm$ 175.52	233.250 $\pm$ 10.02	252.500 $\pm$ 9.82
TP (mg/L)	2.000 $\pm$ 1.83	683.500 $\pm$ 113.49	331.250 $\pm$ 25.75	74.000 $\pm$ 9.32	2.750 $\pm$ 2.06	2.500 $\pm$ 1.08
Chlorophyll- <i>a</i> (mg/L)	0.960 $\pm$ 0.32	2.280 $\pm$ 0.31	15.165 $\pm$ 5.40	32.694 $\pm$ 7.30	0.600 $\pm$ 0.05	0.658 $\pm$ 0.20
Secchi Depth (m)	3.480 $\pm$ 0.42	0.100 $\pm$ 0.00	0.775 $\pm$ 0.21	0.525 $\pm$ 0.09	4.150 $\pm$ 0.73	4.950 $\pm$ 0.35
Trophic Status Index*	28.792	76.566	69.057	66.508	27.882	26.996
TSI Classification*	O	HE	E	E	O	O
Water, filtered						
Mn (μ g/L)	1.810 $\pm$ 1.28	18.145 $\pm$ 7.20	22.175 $\pm$ 4.40	19.768 $\pm$ 8.68	1.948 $\pm$ 0.28	1.325 $\pm$ 0.37
Fe (μ g/L)	24.750 $\pm$ 2.21	556.015 $\pm$ 80.25	314.980 $\pm$ 30.07	128.438 $\pm$ 55.47	27.998 $\pm$ 6.15	32.880 $\pm$ 3.46
Cu (μ g/L)	1.215 $\pm$ 0.14	5.205 $\pm$ 1.48	3.728 $\pm$ 0.28	2.152 $\pm$ 0.08	1.430 $\pm$ 0.41	0.800 $\pm$ 0.04
Zn (μ g/L)	3.030 $\pm$ 0.35	15.240 $\pm$ 2.17	12.570 $\pm$ 3.73	9.485 $\pm$ 1.81	2.775 $\pm$ 0.85	1.915 $\pm$ 0.34
Ba (μ g/L)	270.410 $\pm$ 0.19	268.320 $\pm$ 5.86	278.460 $\pm$ 5.81	273.245 $\pm$ 5.12	257.100 $\pm$ 5.04	230.580 $\pm$ 5.13

Table S2.3 Continued.

Parameter	Upper Lagoon	Lagoon	Finger	Airplane	R1	R2
Pb (µg/L)	0.160 ± 0.03	0.245 ± 0.08	0.205 ± 0.02	0.136 ± 0.02	0.128 ± 0.03	0.158 ± 0.05
THg (ng/L)	2.711 ± 0.08	2.895†	2.343 ± 0.43	1.832 ± 0.17	1.714 ± 0.12	2.139 ± 0.05
MeHg (ng/L)	0.072 ± 0.001	0.252†	0.185 ± 0.002	0.043 ± 0.008	0.061 ± 0.009	0.043 ± 0.004
Sediment						
LOI (loss on ignition) (%)	11.586 ± 0.43	12.879 ± 0.10	16.773 ± 0.31	15.761 ± 0.18	9.676 ± 0.40	12.239 ± 0.29
Mn (µg/g)	277.88 ± 32.03	161.39 ± 1.62	178.30 ± 17.40	233.90 ± 13.93	226.49 ± 26.85	251.69 ± 13.07
Fe (mg/g)	19.246 ± 0.96	19.377 ± 0.08	16.486 ± 2.97	20.059 ± 2.13	18.966 ± 0.77	21.312 ± 3.01
Cu (µg/g)	19.73 ± 4.37	22.16 ± 0.52	41.02 ± 1.56	32.00 ± 0.72	7.396 ± 0.44	8.284 ± 1.23
Zn (µg/g)	65.49 ± 23.78	36.16 ± 0.78	87.30 ± 14.94	72.60 ± 6.10	60.40 ± 2.01	42.49 ± 5.49
Ba (µg/g)	131.60 ± 2.03	148.07 ± 0.08	152.19 ± 1.93	178.6 ± 1.57	127.07 ± 0.50	203.55 ± 3.19
Pb (µg/g)	9.00 ± 1.26	5.46 ± 0.04	7.47 ± 1.20	9.90 ± 0.77	2.880 ± 0.34	6.084 ± 1.49
Sediment [Hg], unadjusted to LOI						
THg (ng/g)	43.392 ± 12.63	29.875 ± 0.80	49.710 ± 9.79	54.037 ± 4.32	27.250 ± 13.71	43.607 ± 13.36
MeHg (ng/g)	0.454 ± 0.18	2.730 ± 0.29	0.866 ± 0.10	0.862 ± 0.13	0.150 ± 0.14	0.543 ± 0.15
Sediment [Hg], adjusted to LOI						
THg (ng/g)	3.745 ± 1.09	2.320 ± 0.06	2.964 ± 0.58	3.428 ± 0.27	2.816 ± 1.42	3.563 ± 1.09
MeHg (ng/g)	0.039 ± 0.02	0.212 ± 0.02	0.052 ± 0.01	0.055 ± 0.01	0.016 ± 0.01	0.044 ± 0.01
Sediment, %MeHg	1.046	9.138	1.742	1.595	0.552	1.245

† Only one replicate.

* Average Trophic Status Index (TSI) calculated from three index variables chlorophyll-*a* (chl_a), Total phosphorous (TP), and Secchi disk (SD.) Equations are as follows:

1) $TSI(chl_a) = 9.81 \ln(chl_a) + 30.6$

2) $TSI(TP) = 14.42 \ln(TP) + 4.15$

3) $TSI(SD) = 60 - 14.41 \ln(SD)$

* TSI Classification: TSI = 0 – 40, oligotrophic (O); 40 – 50, mesotrophic (M), 50 – 70, eutrophic (E), 70 – 80, hypereutrophic (HE).

Method detection limit is 0.30 mg/L selenium (Se).

Table S2.4 Fish species represented in this study, their time of capture, total number captured across sampling sites (n) for those identified as female, male, and immature. Mean size and age (calculated for each lake) are reported with standard deviation. Values are mean $\pm$ standard error.

Scientific name	Common name	Capture month/years	Site-type	Lake	n			Mean size $\pm$ SD	Mean age $\pm$ SD
					Female	Male	Immature		
<i>Thymallus arcticus</i>	Arctic grayling	Jul.-Aug. 2019/2021	Wastewater	Finger	0	4	20	137.583 $\pm$ 14.41	1.000 $\pm$ 0.00
				Airplane	7	9	14	238.400 $\pm$ 92.98	3.241 $\pm$ 1.88
		Jul.-Aug. 2019/2021	Reference	R1	5	7	0	310.917 $\pm$ 33.38	6.700 $\pm$ 1.567
				R2	1	1	1	274.333 $\pm$ 59.28	5.000 $\pm$ 2.65
		Jul.-Aug. 2021	Upstream	Upper Lagoon	6	2	7	255.000 $\pm$ 68.76	5.600 $\pm$ 4.05
<i>Lota lota</i>	Burbot	Jul.-Aug. 2019/2021	Wastewater	Finger	1	2	2	219.800 $\pm$ 91.96	3.636 $\pm$ 0.92
				Airplane	1	5	5	254.909 $\pm$ 47.34	3.000 $\pm$ 1.223
		Jul.-Aug. 2019/2021	Reference	R1	2	5	2	239.444 $\pm$ 47.34	6.222 $\pm$ 2.17
				R2	1	4	0	365.400 $\pm$ 63.13	9.000 $\pm$ 1.41
		Jul.-Aug. 2021	Upstream	Upper Lagoon	1	0	0	255.000 $\pm$ 0.00	7.000 $\pm$ 0.00

Table S2.5 Summary of data from biota samples in six lakes of Baker Lake, Nunavut, Canada. Upper Lagoon Lake is situated upstream of the wastewater wetland, while Lagoon Lake, Finger Lake, and Airplane constitute the lagoon-tundra wastewater wetland. Lake R1 and Lake R2 serve as reference systems. Analyses were conducted on bulk seston, and the muscle and liver tissues of Arctic grayling (*Thymallus arcticus*) and Burbot (*Lota lota*). Biotic samples were not collected in Lagoon Lake due to feasibility concerns. Values are mean dry weight $\pm$ standard error.

Parameter	Finger	Airplane	R1	R2
Periphyton, n	1	1	1	1
d ¹³ C (‰)	-17.580	-12.410	-21.850	-18.450
d ¹⁵ N (‰)	11.950	8.420	0.990	0.960
Bulk seston, n	2	2	2	2
Mn (µg/g)	257.243 $\pm$ 4.46	103.477 $\pm$ 6.07	187.453 $\pm$ 3.07	258.579 $\pm$ 6.19
Fe (µg/g)	6057.262 $\pm$ 37.79	3105.612 $\pm$ 25.22	2744.350 $\pm$ 24.43	4928.714 $\pm$ 36.22
Cu (µg/g)	33.070 $\pm$ 1.87	22.025 $\pm$ 1.87	21.723 $\pm$ 1.90	18.316 $\pm$ 1.76
Zn (µg/g)	119.336 $\pm$ 3.78	95.543 $\pm$ 2.07	130.559 $\pm$ 3.28	107.446 $\pm$ 1.12
Ba (µg/g)	97.681 $\pm$ 3.65	41.526 $\pm$ 2.84	40.268 $\pm$ 2.90	53.994 $\pm$ 1.93
Pb (µg/g)	12.367 $\pm$ 1.33	10.666 $\pm$ 1.04	16.430 $\pm$ 0.42	9.51 $\pm$ 0.54
THg (µg/g)	0.106 [†]	0.042 [†]	0.205 [†]	0.164 $\pm$ 0.02
MeHg (µg/g)	0.082 [†]	0.021 [†]	0.083 [†]	0.039 [†]
d ¹³ C (‰)	-23.770 [†]	-26.690 [†]	-28.980 [†]	-29.790 [†]
d ¹⁵ N (‰)	9.030 [†]	11.950 [†]	5.470 [†]	5.570 [†]
Arctic grayling (<i>Thymallus arcticus</i>)				
n, muscle tissue	23	29	14	3
Mn (µg/g)	0.718 $\pm$ 0.02	0.645 $\pm$ 0.02	1.527 $\pm$ 0.08	0.831 $\pm$ 0.22
Fe (µg/g)	18.033 $\pm$ 0.12	17.870 $\pm$ 0.09	24.386 $\pm$ 0.37	19.586 $\pm$ 0.85
Cu (µg/g)	1.572 $\pm$ 0.02	1.592 $\pm$ 0.02	<MRL*	1.054 $\pm$ 1.41
Zn (µg/g)	28.545 $\pm$ 0.11	18.614 $\pm$ 0.07	19.890 $\pm$ 0.26	20.432 $\pm$ 0.41
Ba (µg/g)	0.231 $\pm$ 0.02	0.783 $\pm$ 0.04	0.537 $\pm$ 0.05	0.604 $\pm$ 0.28
Pb (µg/g)	0.20 $\pm$ 0.00	<MRL	<MRL	<MRL
THg (µg/g)	0.161 $\pm$ 0.01	0.146 $\pm$ 0.01	0.614 $\pm$ 0.05	0.657 $\pm$ 0.10
MeHg (µg/g)	0.154 $\pm$ 0.01	0.131 $\pm$ 0.01	0.551 $\pm$ 0.05	0.649 $\pm$ 0.08
%MeHg	95.790 $\pm$ 0.14	89.099 $\pm$ 0.13	91.204 $\pm$ 0.29	99.309 $\pm$ 0.83
d ¹³ C (‰)	-19.734 $\pm$ 0.03	-20.324 $\pm$ 0.03	-21.216 $\pm$ 0.07	-23.098 $\pm$ 0.31
d ¹⁵ N (‰)	14.438 $\pm$ 0.03	13.192 $\pm$ 0.03	10.391 $\pm$ 0.05	8.052 $\pm$ 0.21
d ¹⁵ N _{adj} (‰)	2.702 $\pm$ 0.03	3.948 $\pm$ 0.03	4.769 $\pm$ 0.05	2.482 $\pm$ 0.21
C:N	3.604 $\pm$ 0.02	3.556 $\pm$ 0.01	3.286 $\pm$ 0.04	3.343 $\pm$ 0.08
n, liver	23	29	9	3
Mn (µg/g)	3.068 $\pm$ 0.07	3.134 $\pm$ 0.05	7.428 $\pm$ 0.16	1.974 $\pm$ 0.25
Fe (µg/g)	259.825 $\pm$ 1.07	154.459 $\pm$ 0.33	705.682 $\pm$ 3.56	76.430 $\pm$ 1.29
Cu (µg/g)	5.290 $\pm$ 0.09	5.096 $\pm$ 0.06	22.784 $\pm$ 0.51	2.986 $\pm$ 0.43
Zn (µg/g)	50.223 $\pm$ 0.26	43.474 $\pm$ 0.18	87.845 $\pm$ 0.44	22.058 $\pm$ 0.52
Ba (µg/g)	0.211 $\pm$ 0.02	0.404 $\pm$ 0.03	0.737 $\pm$ 0.12	0.113 $\pm$ 0.06
Pb (µg/g)	0.390 $\pm$ 0.21	0.040 [†]	0.018 [†]	<MRL

Table S2.5 Continued.

Parameter	Finger	Airplane	R1	R2
THg (µg/g)	0.199 ± 0.02	0.144 ± 0.06	0.669 ± 0.06	0.173 ± 0.02
MeHg (µg/g)	0.188 ± 0.02	0.093 ± 0.01	0.517 ± 0.07	0.178 ± 0.04
Burbot (<i>Lota lota</i>)				
n, muscle tissue	11	5	9	4
Mn (µg/g)	1.740 ± 0.17	1.879 ± 0.08	2.310 ± 0.11	1.556 ± 0.15
Fe (µg/g)	24.253 ± 0.65	32.137 ± 0.42	25.537 ± 0.35	22.038 ± 1.03
Cu (µg/g)	2.409†	<MRL	1.500†	1.478†
Zn (µg/g)	20.734 ± 0.37	19.445 ± 0.17	18.168 ± 0.18	15.220 ± 0.26
Ba (µg/g)	1.311 ± 0.22	1.493 ± 0.10	1.560 ± 0.12	0.270 ± 0.06
Pb (µg/g)	<MRL	<MRL	<MRL	0.035†
THg (µg/g)	0.289 ± 0.11	0.264 ± 0.03	1.097 ± 0.08	1.321 ± 0.20
MeHg (µg/g)	0.278 ± 0.11	0.278 ± 0.11	1.005 ± 0.07	1.117 ± 0.18
%MeHg	95.758 ± 0.13	89.236 ± 0.14	89.958 ± 0.29	99.309 ± 0.83
δ ¹³ C (‰)	-19.580 ± 0.17	-19.410 ± 0.09	-20.099 ± 0.12	-21.269 ± 0.12
δ ¹⁵ N (‰)	14.964 ± 0.18	15.109 ± 0.07	11.190 ± 0.09	11.813 ± 0.21
δ ¹⁵ N _{adj} (‰)	4.474 ± 0.18	4.619 ± 0.07	5.760 ± 0.09	6.383 ± 0.21
C:N	3.382 ± 0.06	3.342 ± 0.02	3.251 ± 0.02	3.254 ± 0.06
n, liver	11	5	9	4
Mn (µg/g)	3.091 ± 0.14	1.791 ± 0.20	7.795 ± 0.31	5.033 ± 0.43
Fe (µg/g)	168.590 ± 0.19	709.153 ± 3.97	443.932 ± 1.80	1250.705 ± 8.38
Cu (µg/g)	18.198 ± 0.36	10.270 ± 0.47	51.619 ± 0.79	214.317 ± 3.44
Zn (µg/g)	51.0711 ± 0.54	25.995 ± 0.55	99.218 ± 0.77	154.531 ± 2.47
Ba (µg/g)	0.243 ± 0.08	0.218 ± 0.03	0.783 ± 0.12	0.406 ± 0.14
Pb (µg/g)	<MRL	0.067 ± 0.07	0.180†	0.080†
THg (µg/g)	0.039 ± 0.03	0.033 ± 0.01	0.371 ± 0.06	0.492 ± 0.17
MeHg (µg/g)	0.027 ± 0.03	0.029 ± 0.01	0.219 ± 0.05	0.217 ± 0.10

* MRL; method reporting limit.

† Only one replicate available for analysis.

Table S2.6 Output from the principal component analysis conducted on water quality parameters. Cos2 represents the quality of representation for variables on the factor map. It is calculated as the squared coordinates. A high cos2 indicates a good representation of the variable on the principal component – meaning the variable is close to the circumference. If the cos2 is low, then the variable is not well represented and will be positioned close to the origin.

Parameter	Unit	Dim.1	Dim.2	cos ²	Contribution (%)
Lead (Pb)	(ms/cm)	0.962	-0.274	1.000	6.416
Total phosphorous (TP)	(mg/L)	0.952	0.294	0.992	6.365
Iron (Fe)	(mg/L)	0.973	0.206	0.990	6.354
Copper (Cu)	(mg/L)	0.985	0.088	0.978	6.276
Manganese (Mn)	(mg/L)	0.978	-0.103	0.968	6.212
Dissolved oxygen (DO)	(mg/L)	-0.798	-0.569	0.960	6.161
Conductivity (Cond)	(ms/cm)	0.939	-0.273	0.957	6.141
Methylmercury (MeHg)	(ng/L)	0.940	0.266	0.954	6.124
Total nitrogen (TN)	(mg/L)	0.912	0.345	0.950	6.097
Dissolved organic carbon (DOC)		0.522	-0.813	0.933	5.987
Total mercury (THg)	(ng/L)	0.914	0.298	0.924	5.934
Zinc (Zn)	(mg/L)	-0.949	-0.123	0.915	5.875
Chlorophyll- <i>a</i>	(mg/L)	0.144	-0.934	0.893	5.729
pH		-0.866	-0.066	0.754	4.842
Sulfate SO ₄	(mg/L)	0.515	-0.586	0.609	3.908
Barium (Ba)	(mg/L)	0.616	-0.479	0.608	3.903
Chlorine (Cl)	(mg/L)	0.537	-0.450	0.491	3.151
Calcium (Ca)	(mg/L)	0.325	-0.510	0.365	2.345
Dissolved inorganic carbon(DIC)		0.309	0.494	0.340	2.179

Table S2.7 Output from the principal component analysis conducted on sediment quality parameters. Cos2 represents the quality of representation for variables on the factor map. It is calculated as the squared coordinates. A high cos2 indicates a good representation of the variable on the principal component – meaning the variable is close to the circumference. If the cos2 is low, then the variable is not well represented and will be positioned close to the origin.

Parameter	Unit	Dim.1	Dim.2	cos ²	Contribution (%)
Cu	(mg/L)	-0.6277806	-0.774914	0.99460022	13.9736377
Mn	(mg/L)	0.9687797	-0.2098431	0.98256824	13.8045943
Zn	(mg/L)	-0.2591351	-0.9509421	0.97144192	13.648275
Pb	(mg/L)	0.75799957	-0.590087	0.922766	12.9644026
MeHg	(ng/L)	-0.6369704	0.70541224	0.90333775	12.6914454
Fe	(mg/L)	0.77585741	0.46636273	0.81944891	11.512849
THg	(ng/L)	0.8382392	-0.1435589	0.72325411	10.16136
LOI		-0.3745533	-0.5147015	0.40520779	5.69296756
Ba	(mg/L)	0.6190777	-0.1086643	0.39506514	5.55046844

Supplementary information for Chapter 3

Table S3. 1 Typical operating conditions for the LA-ICP-MS (University of Manitoba).

Laser conditions		ICP-MS conditions		Data acquisition	
Repetition	10 Hz	Plasma power	1238 (W)	Protocol	Time resolved analysis
Spot size	30 μm	Cool Gas	15.6 (L/min)	Scanning mode	BScan and Escan
Laser scan	2 $\mu\text{m/s}$	Auxiliary gas	1.00 (L/min)	Detector mode	Analog and counting
Power	80%	Sample gas (AR)	0.82 (L/min)	Dwell time	0.001 – 0.3 ms
Incident pulse	0.01 mJ	Additional gas (HE)	0.80 (L/min)		

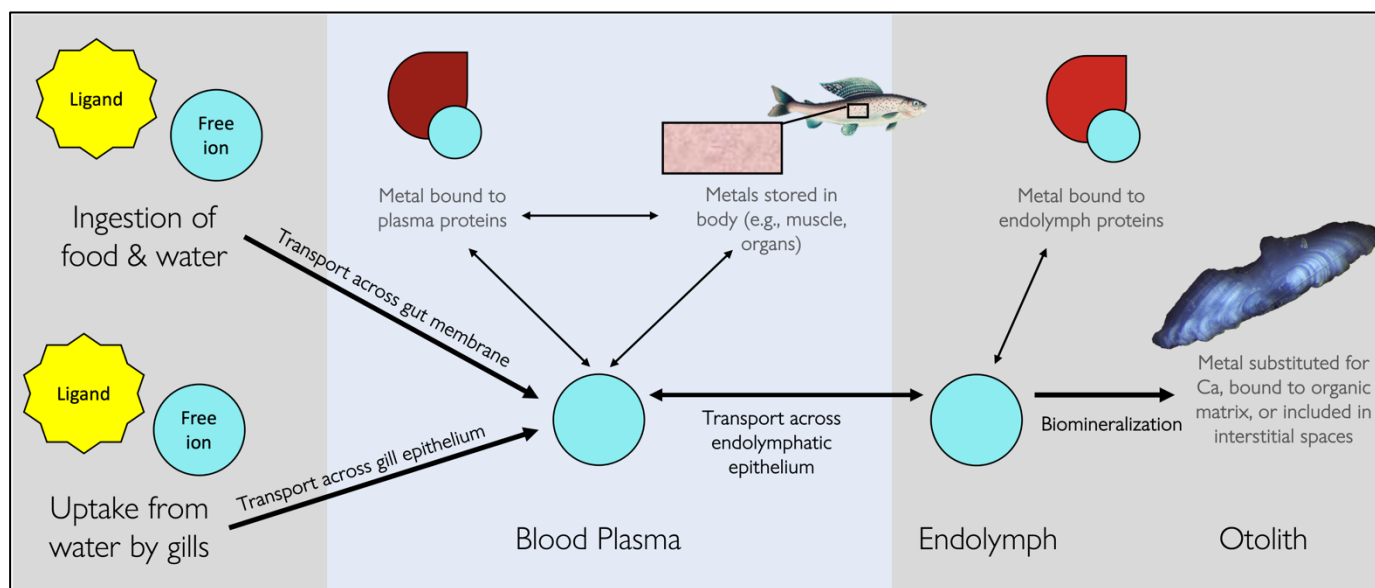


Figure S3.1 Schematic modified from Campana et al. (1999) and Hüssy et al. (2021).

Table S3.2 Pearson correlation coefficients between metal concentrations in marginal edge of otoliths and soft-tissue concentrations (i.e., muscle, liver) from fish captured in Airplane Lake. Individual fish were treated as replicates (n = 29). Significant relationships (p < 0.05) are underlined and bold.

	Annulus width	LogMn otolith	LogFe otolith	LogCu otolith	LogZn otolith	LogPb otolith	LogMn tissue	LogFe tissue	LogZn tissue	LogMn liver	LogFe liver	LogCu liver	LogZn liver
Annulus width													
LogMn otolith	<u>0.423</u>												
LogFe otolith	-0.225	<u>-0.463</u>											
LogCu otolith	0.152	<u>0.385</u>	-0.014										
LogZn otolith	0.312	<u>0.634</u>	<u>-0.436</u>	0.185									
LogPb otolith	-0.226	0.166	-0.315	<u>0.464</u>	0.103								
LogMn tissue	0.065	-0.064	-0.024	0.104	-0.134	-0.096							
LogFe tissue	0.012	-0.123	-0.023	0.180	0.201	0.098	0.054						
LogZn tissue	-0.156	0.109	-0.146	<u>0.542</u>	0.348	<u>0.629</u>	-0.197	0.317					
LogMn liver	-0.056	0.121	-0.049	0.155	0.070	0.212	-0.106	-0.145	0.126				
LogFe liver	0.207	0.209	0.026	0.075	0.138	0.136	-0.219	-0.368	-0.159	<u>0.540</u>			
LogCu liver	-0.180	-0.318	0.081	0.357	-0.196	0.028	-0.088	<u>0.429</u>	0.232	0.048	-0.178		
LogZn liver	-0.201	-0.225	0.208	<u>0.480</u>	-0.207	0.165	0.031	0.218	0.386	<u>0.450</u>	0.090	<u>0.662</u>	

Table S3.3 Pearson correlation coefficients between fish life-history characteristics and marginal edge otolith metal concentrations in fish from Airplane Lake. Individual fish were treated as replicates (n = 29). Significant relationships (p < 0.05) are underlined and bold.

	Age	Length	Weight	LogK	LogLSI	GSI	Growth rate	N15adj	C13cor	LogCN	Annulus width	LogMn otolith	LogFe otolith	LogCu otolith	LogZn otolith	LogPb otolith
Age																
Length	<u>0.914</u>															
Weight	<u>0.920</u>	<u>0.966</u>														
LogK	0.327	<u>0.438</u>	<u>0.463</u>													
LogLSI	0.317	0.313	0.356	0.357												
GSI	<u>0.639</u>	<u>0.651</u>	<u>0.738</u>	0.375	0.317											
Growth rate	<u>-0.923</u>	<u>-0.806</u>	<u>-0.794</u>	-0.265	-0.303	<u>-0.498</u>										
N15adj	-0.294	-0.290	-0.223	0.453	0.243	0.030	<u>0.379</u>									
C13cor	0.046	0.018	-0.066	-0.388	<u>-0.430</u>	-0.319	0.094	-0.192								
LogCN	0.266	0.275	0.252	-0.252	-0.033	0.118	-0.409	<u>-0.884</u>	-0.287							
Annulus width	-0.285	<u>-0.398</u>	<u>-0.372</u>	-0.157	0.161	-0.342	0.231	0.126	-0.085	-0.079						
LogMn otolith	<u>-0.590</u>	<u>-0.656</u>	<u>-0.712</u>	<u>-0.377</u>	-0.095	-0.388	<u>0.523</u>	0.174	-0.002	-0.173	<u>0.423</u>					
LogFe otolith	<u>0.375</u>	<u>0.426</u>	<u>0.466</u>	0.261	-0.059	0.300	-0.292	-0.410	0.120	0.344	-0.225	<u>-0.463</u>				
LogCu otolith	-0.220	-0.247	-0.280	-0.156	-0.005	-0.287	0.190	-0.140	0.085	0.100	0.152	<u>0.385</u>	-0.014			
LogZn otolith	<u>-0.619</u>	<u>-0.695</u>	<u>-0.726</u>	-0.256	-0.092	<u>-0.649</u>	0.445	0.223	-0.172	-0.136	0.312	<u>0.634</u>	<u>-0.436</u>	0.185		
LogPb otolith	-0.177	-0.126	-0.227	-0.197	-0.198	-0.187	0.033	-0.355	-0.019	0.352	-0.226	0.166	-0.315	<u>0.464</u>	0.103	

Table S3.4 Pearson correlation coefficients between fish marginal edge otolith metal concentrations and soft-tissue concentrations (i.e., muscle, liver) in Lake R1. Individual fish were treated as replicates (n = 14). Significant relationships (p < 0.05) are underlined and bold.

	Annulus width	LogMn otolith	LogFe otolith	LogCu otolith	LogZn otolith	LogPb otolith	LogMn tissue	LogFe tissue	LogZn tissue	LogMn liver	LogFe liver	LogCu liver	LogZn liver
Annulus width													
LogMn otolith	0.137												
LogFe otolith	-0.526	-0.165											
LogCu otolith	-0.376	0.154	0.147										
LogZn otolith	0.090	<u>0.573</u>	-0.153	0.023									
LogPb otolith	-0.471	0.289	0.195	<u>0.803</u>	-0.132								
LogMn tissue	0.101	0.037	0.213	0.013	-0.494	0.183							
LogFe tissue	-0.209	-0.305	0.246	0.215	-0.260	0.049	0.526						
LogZn tissue	-0.143	-0.380	-0.179	-0.065	-0.165	-0.288	0.173	0.374					
LogMn liver	0.218	0.323	-0.052	0.348	-0.200	0.554	0.551	0.098	0.065				
LogFe liver	-0.008	0.041	-0.223	0.281	-0.085	0.267	0.038	0.286	0.434	<u>0.597</u>			
LogCu liver	-0.334	-0.201	0.135	-0.230	-0.342	-0.038	0.027	0.166	-0.280	-0.396	-0.238		
LogZn liver	-0.262	-0.196	-0.074	0.162	<u>-0.583</u>	0.229	0.171	0.343	0.094	0.096	0.335	<u>0.709</u>	

Table S3.5 Pearson correlation coefficients between fish life-history characteristics and marginal edge otolith metal concentrations in Lake R1. Individual fish were treated as replicates (n = 13). Significant relationships (p < 0.05) are underlined and bold.

	Age	Length	Weight	LogK	LogLSI	GSI	Growth rate	N15adj	C13cor	LogCN	Annulus width	LogMn otolith	LogFe otolith	LogCu otolith	LogZn otolith	LogPb otolith
Age																
Length	<u>0.650</u>															
Weight	0.525	<u>0.984</u>														
LogK	<u>-0.781</u>	<u>-0.911</u>	<u>-0.835</u>													
LogLSI	<u>0.501</u>	0.039	-0.068	-0.192												
GSI	0.295	0.307	0.278	-0.290	0.511											
Growth rate	<u>-0.909</u>	-0.361	-0.234	0.475	<u>-0.590</u>	-0.289										
N15adj	0.454	<u>0.658</u>	<u>0.661</u>	-0.533	-0.111	-0.174	-0.264									
C13cor	0.472	0.460	0.449	-0.299	0.147	0.202	-0.432	<u>0.587</u>								
LogCN	-0.496	<u>-0.665</u>	<u>-0.666</u>	0.515	0.056	0.089	0.330	<u>-0.976</u>	<u>-0.748</u>							
Annulus width	-0.341	-0.030	0.064	0.139	-0.174	0.054	0.339	-0.148	-0.483	0.246						
LogMn otolith	-0.163	-0.380	-0.388	0.253	-0.010	<u>-0.747</u>	0.154	0.104	-0.209	-0.026	0.137					
LogFe otolith	<u>0.528</u>	0.390	0.304	-0.391	0.532	0.186	-0.482	0.249	<u>0.537</u>	-0.347	-0.526	-0.165				
LogCu otolith	0.209	0.101	0.016	-0.308	0.376	-0.094	-0.057	0.007	-0.285	0.072	-0.376	0.154	0.147			
LogZn otolith	<u>-0.483</u>	<u>-0.550</u>	<u>-0.561</u>	0.359	-0.144	-0.481	0.512	-0.364	-0.472	0.430	0.090	<u>0.573</u>	-0.153	0.023		
LogPb otolith	0.385	0.215	0.139	-0.364	0.441	-0.094	-0.210	0.321	0.196	-0.311	-0.471	0.289	0.195	<u>0.803</u>	-0.132	

Table S3.6 Pearson correlation coefficients between fish marginal edge otolith metal concentrations and soft-tissue concentrations (i.e., muscle, liver) in fish from Upper Lagoon Lake. Individual fish were treated as replicates (n = 14). Significant relationships (p < 0.05) are underlined and bold.

	Annulus width	LogMn otolith	LogFe otolith	LogCu otolith	LogZn otolith	LogPb otolith	LogMn tissue	LogFe tissue	LogZn tissue	LogMn liver	LogFe liver	LogCu liver	LogZn liver
Annulus width													
LogMn otolith	<u>0.547</u>												
LogFe otolith	0.336	0.307											
LogCu otolith	0.132	0.483	0.111										
LogZn otolith	<u>0.715</u>	<u>0.768</u>	0.250	0.205									
LogPb otolith	<u>0.542</u>	0.257	<u>0.605</u>	0.454	0.284								
LogMn tissue	-0.029	-0.011	-0.046	-0.136	0.195	0.122							
LogFe tissue	-0.282	-0.068	-0.186	-0.038	-0.303	-0.282	-0.146						
LogZn tissue	<u>0.752</u>	0.169	-0.222	-0.175	0.381	0.249	0.119	-0.057					
LogMn liver	0.327	-0.185	-0.100	-0.458	0.351	-0.026	0.098	<u>-0.706</u>	0.430				
LogFe liver	0.058	-0.527	-0.197	-0.425	-0.012	-0.151	0.351	-0.512	0.226	<u>0.669</u>			
LogCu liver	-0.094	-0.466	-0.270	-0.057	-0.059	-0.214	-0.098	-0.504	-0.071	0.502	<u>0.758</u>		
LogZn liver	0.065	-0.451	-0.289	-0.277	0.112	-0.090	0.078	<u>-0.651</u>	0.225	<u>0.809</u>	<u>0.788</u>	<u>0.875</u>	

Table S3.7 Pearson correlation coefficients between fish life-history characteristics and marginal edge otolith metal concentrations in fish from Upper Lagoon Lake. Individual fish were treated as replicates (n = 14). Significant relationships (p < 0.05) are underlined and bold.

	Age	Length	Weight	LogK	LogLSI	GSI	Growth rate	N15adj	C13cor	LogCN	Annulus width	LogMn otolith	LogFe otolith	LogCu otolith	LogZn otolith	LogPb otolith
Age																
Length	<u>0.685</u>															
Weight	<u>0.582</u>	<u>0.987</u>														
LogK	<u>-0.661</u>	-0.447	-0.334													
LogLSI	0.405	0.146	0.088	-0.304												
GSI	0.340	0.567	0.482	-0.365	0.449											
Growth rate	<u>-0.930</u>	-0.842	-0.780	0.460	-0.364	-0.392										
N15adj	<u>0.587</u>	0.060	-0.067	-0.636	0.184	0.310	-0.308									
C13cor	-0.354	0.303	0.384	0.337	-0.201	-0.429	0.064	<u>-0.622</u>								
LogCN	-0.528	-0.227	-0.116	0.611	-0.116	-0.255	0.334	<u>-0.913</u>	0.253							
Annulus width	<u>-0.785</u>	<u>-0.636</u>	<u>-0.548</u>	0.408	-0.506	0.081	<u>0.822</u>	-0.368	-0.068	0.464						
LogMn otolith	<u>-0.583</u>	<u>-0.667</u>	<u>-0.617</u>	<u>0.605</u>	-0.474	-0.397	<u>0.631</u>	-0.132	-0.063	0.212	<u>0.547</u>					
LogFe otolith	-0.265	0.056	0.119	0.192	-0.313	0.301	0.210	0.078	0.250	-0.246	0.336	0.307				
LogCu otolith	-0.179	-0.332	-0.352	0.117	0.195	-0.282	0.283	0.074	-0.047	-0.042	0.132	0.483	0.111			
LogZn otolith	<u>-0.869</u>	<u>-0.678</u>	<u>-0.587</u>	<u>0.704</u>	<u>-0.618</u>	-0.573	<u>0.826</u>	-0.462	0.174	0.479	<u>0.715</u>	<u>0.768</u>	0.250	0.205		
LogPb otolith	-0.473	-0.328	-0.273	0.119	0.085	0.547	<u>0.551</u>	-0.002	-0.082	0.023	<u>0.542</u>	0.257	<u>0.605</u>	0.454	0.284	

Table S3.8 Results from analysis of Variance (ANOVA) of year-one and marginal edge otolith metal concentrations. Pairwise comparisons are computed with Tukey's Honestly Significant Difference (HSD) with *p*-values adjusted for multiple comparisons.

Otolith Region	Metal (ppm)	ANOVA Results		Tukey's HSD Results			
		F	df	p	UL-AIR	R1-AIR	UL-R1
Year-one	Mn	2.359	2, 48	0.105	0.124	0.278	0.915
	Fe	5.090	2, 48	<u>0.010</u>	<u>0.009</u>	0.060	0.872
	Cu	4.914	2, 48	<u>0.011</u>	0.459	0.077	<u>0.010</u>
	Zn	7.515	2, 48	<u>0.001</u>	0.090	0.087	<u><0.001</u>
	Pb	0.478	2, 48	0.623	0.932	0.594	0.843
[Length-adjusted] marginal edge	Mn	1.345	2, 50	0.270	0.640	0.550	0.245
	Fe	8.214	2, 50	<u><0.001</u>	<u>0.001</u>	<u>0.043</u>	0.719
	Cu	3.281	2, 50	<u>0.046</u>	0.083	0.148	0.999
	Zn	4.080	2, 50	<u>0.023</u>	0.798	<u>0.017</u>	0.118
	Pb	1.870	2, 50	0.165	0.174	0.484	0.914

Table S3.9. Output from the principal component analysis conducted on first year otolith metal concentrations. Cos2 represents the quality of representation for variables on the factor map. It is calculated as the squared coordinates. A high cos2 indicates a good representation of the variable on the principal component – meaning the variable is close to the circumference. If the cos2 is low, then the variable is not well represented and will be positioned close to the origin.

Parameter	Dim.1	Dim.2	cos ²	Contribution (%)
Lead (Pb)	0.402	0.771	0.757	24.065
Copper (Cu)	0.831	0.193	0.727	23.118
Zinc (Zn)	0.665	-0.436	0.633	20.114
Iron (Fe)	-0.541	0.578	0.627	19.947
Manganese (Mn)	0.593	0.223	0.401	12.755

Table S3.10 Output from the principal component analysis conducted on marginal edge otolith metal concentrations. Cos2 represents the quality of representation for variables on the factor map. It is calculated as the squared coordinates. A high cos2 indicates a good representation of the variable on the principal component – meaning the variable is close to the circumference. If the cos2 is low, then the variable is not well represented and will be positioned close to the origin.

Parameter	Dim.1	Dim.2	cos²	Contribution (%)
Lead (Pb)	-0.830	0.352	0.813	23.697
Zinc (Zn)	-0.332	-0.765	0.696	20.306
Manganese (Mn)	-0.399	-0.716	0.673	19.610
Copper (Cu)	-0.809	0.120	0.669	19.505
Iron (Fe)	-0.732	0.207	0.579	16.882

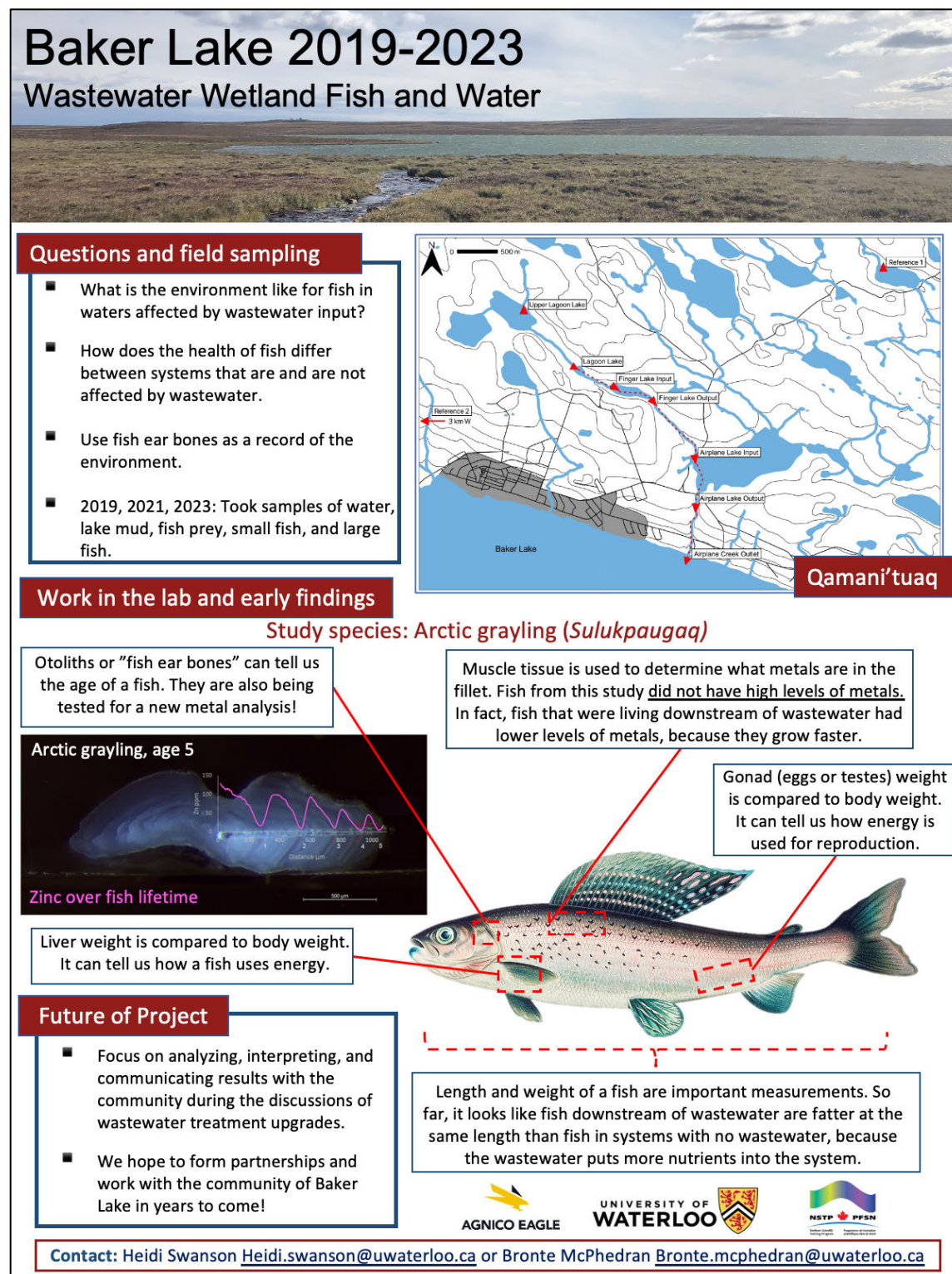


Figure S4.1 A poster was used to share project findings with residents of Baker Lake, NU. An information booth was set up at the local grocery store to encourage questions and discussion.

