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Spatiotemporal changes in feeding ecology of brook trout in sub-alpine lakes at Gaspésie National Park

Master's thesis in biology

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Abstract

As a result of strong anthropogenic impacts such as climate change, invasive species and extreme weather, freshwater ecosystems are facing global degradation. Due to these effects the understanding of a changing environment is being more and more frequently studied in order to cope with such changes. A central part of understanding species reaction toward environmental changes is by studying the feeding ecology which provides many aspects of trophic interactions. The main objective in this research is to assess spatial and temporal changes in the feeding ecology of brook trout (*Salvelinus fontinalis*) inhabiting alpine lakes via stomach content analysis. Because feeding strategy for opportunistic feeders like brook trout was long believed to not practice individual specialization, this study aims to dig deeper into the examination of individual specialization within generalist populations. The feeding ecology of brook trout was investigated with various indices related to diet overlap, individual specialization, and species diversity. This research also estimates the relative importance for all prey items as well as group them in the following prey groups: fish, zooplankton, terrestrial invertebrates, and benthic macroinvertebrates. The general pattern, show that the different populations did not exhibit either strong specialization or strong generalization in feeding strategy among brook trout populations. However, some populations expressed seasonal variation indicating that they indeed were individual specialists among the generalist population and vice versa. This research suggested that the most explanatory reason for some prey items being highly represented in their diet was because of prey availability and abundance. This research has contributed to a broader understanding of freshwater ecosystem dynamics in alpine lakes. Additionally, the results from this research can be used to inform conservation efforts, aiming to preserve biodiversity and ecosystem resilience in the face of global environmental changes.

Preferences

This master thesis is submitted for the Faculty of Environmental Sciences and Natural Resource Management (MINA) at the Norwegian University of Life Sciences (NMBU), as part of the course M60-ECOL, master's thesis. This research is weighted 60 ECTS credits and marks the completion of my master's degree of biology. Research project has been performed under the supervision of Associate Professor Louise CHavarie and Leah Strople during the summer 2023.

Table of content

<i>Abstract</i>	2
<i>Preferences</i>	3
<i>1. Introduction</i>	5
<i>2. Materials and methods</i>	7
2.1. Study area	7
2.2. Field sampling	8
2.3. Laboratory work	9
2.4. Relative importance index	10
2.5. Stomach content analysis	11
2.5.1. Diet overlap and individual specialization	11
2.5.2. Diversity index	13
2.5.3. Generalized Linear Models (GLM)	14
3. Results	15
3.1. Relative importance	15
3.2. Dietary analysis	18
3.3. GLM and GLMM	22
4. Discussion	24
4.1 Relative Importance Index	24
4.2 Stomach content analysis	24
4.3 GLM and GLMM	26
4.4 Limitations of this study	26
5. Conclusion	27
<i>References</i>	28

1. Introduction

Freshwater ecosystems are among the most extensively altered ecosystems on the planet, experiencing changes due to various anthropogenic impacts, such as climate change, the introduction of new invasive species, and extreme weather events like floods and draught (Dudgeon et al., 2006; Reid et al., 2019). These impacts are affecting distribution and interactions among species and are highly dependent on the temporal and spatial scales (Carpenter et al., 1992, 2011). Furthermore, freshwater ecosystems such as sub-alpine and alpine lakes, undergo strong seasonal variability with year-to-year fluctuations due to large abiotic and biotic changes in the environment (Sotiropoulos et al., 2006). Additionally, sub-alpine and alpine lakes, characterized by their simple structure and low productivity, are particularly vulnerable towards any environmental variation (Tiberti et al., 2016). In these type of lakes, salmonid fish species is often the sole fish species present (Tiberti et al., 2016), and even though salmonid species are highly plastic (Behnke, 2002), previous research has shown that salmonid species are susceptible to degradation from anthropogenic impacts (Amundsen & Klemetsen, 1988).

Understanding predator-prey relationships and food web structure through high-quality research is considered essential for comprehending ecological interactions among species (Mizsei et al., 2019). Stomach content analysis in fish species serve as a common method to study foraging strategies within and among populations, especially focusing on intra- and interspecific competition, providing valuable insights into feeding ecology, habitat use, and nutrient flow in aquatic ecosystems (Braga et al., 2012; Manko, 2016; Amundsen & Sánchez-Hernández, 2019; Haque et al., 2021). Fish are a particularly suitable group of organisms for dietary studies as they can often be sampled in high numbers and typically swallow their prey whole, making stomach content identification easier (Amundsen & Sánchez-Hernández, 2019). While stomach content analysis offers a unique snapshot of the fish diet, providing very precise information about foraging strategy for a specific times and places, it may not capture the long-term feeding strategies and can easily be biased by factors such as regurgitation during capture, identification of partial prey items or/and underestimation of prey size (Vinson & Budy, 2011; Chavarie et al., 2014). Additionally, stomach content analysis has some major disadvantages where the contribution of slowly digested prey taxa tends to be overestimated (Amundsen & Sánchez-Hernández, 2019), and the methodology

related to mass or volume measurements can be very time consuming (Hyslop, 1980; Baker et al., 2014).

Brook trout (*Salvelinus fontinalis*) are vital inhabitants of many sub-alpine lakes in North America. With their remarkable capacity for plasticity (Behnke, 2002) brook trout often play an integral role as the top predator in sub-alpine lakes (Baum & Worm, 2009). They are also the most warm-tolerant species in the *Salvelinus* genus (Behnke, 2002), serving as an ideal sentinel species for understanding the influence of climate change on cold-water fishes (Warren et al., 2012, 2017; Bassar et al., 2016; Jirka & Kraft, 2017). Unfortunately, the native range of brook trout in North America, especially in the United States, has been significantly reduced (Behnke, 2002), emphasizing the need for detailed knowledge of their ecology for optimal understanding of their response to a changing environment (McFadden, 1961).

Brook trout are recognized as strong opportunistic feeders, primarily consuming aquatic and terrestrial invertebrates, followed by fishes and amphibians (Behnke, 2002; Jirka & Kraft, 2017). Initially, generalistic feeding strategies were thought to be prevalent among brook trout individuals (Allan, 1978; Lomnicki, 1988). However, previous studies defining generalist populations overlooked the possibility of strong individual specialization within generalist populations (Feinsinger et al., 1981; Linton et al., 1981), despite the fact that ecologists have known for a long time that individuals within a population can differ substantially in their resource use, expressing strong individual specialization (Van Valen, 1965). More recent research has shed light on the prevalence of individual specialization, particularly in response to variations in prey abundance and seasonal changes, suggesting that individual specialization is maybe more common than previously thought (Bolnick et al., 2002, 2003, 2004; Araújo et al., 2008; Hulsman et al., 2016; Tiberti et al., 2016; Haque et al., 2021). Furthermore, diet composition has also shown to be strongly affected by intra- and interspecific competition in streams where brook trout live sympatrically with other fish species (Magnan & Don Stevens, 1993; Browne & Rasmussen, 2009). However, fewer studies have investigated the diet composition of brook trout in mountain regions in their native range, particularly focusing on seasonal and spatial variation, and intra- and interspecific competition with their closest relatives in the *Salvelinus* genus. The primary objective in this research is therefore to assess spatial and temporal changes in the feeding ecology of brook trout (*Salvelinus fontinalis*) inhabiting alpine lakes via stomach content analysis. This study aims to: (1) examine seasonal variations in brook trout diet between

summer and winter, (2) quantify brook trout diet differences among lakes sustaining different *Salvelinus* assemblages, (3) determine generalist-specialist feeding tactics of alpine brook trout populations, and 4) assess if physiological **traits** differences lead to variations in diet.

2. Materials and methods

2.1. Study area

This study was conducted in the southeastern region of Québec (Canada; ~ 48°56'N, 66°14'W), within the Gaspésie National Park (*Figure 1*). The four studied lakes are situated at altitudes ranging from 469 meters to 683 meters above sea level and are characterized as sub-alpine lakes. Typically, the lakes are covered with ice from late November until late May, although there is large year-to-year fluctuation (M. L'Italien, *personal communication*, 28.02.2024). The selected lakes of the study vary in some physical characteristics, both in lake depth and surface area, with Lake Haymard being the smallest (8 ha) with deepest point only being 3.0 meters, while Lake Cascapédia is the largest (129 ha) where the deepest point being 18.3 meters (*Table 1*). All lakes, with the exception of Cascapédia, are protected against recreational fishing and undergo minimal anthropogenic disturbances. Lake Paul and Lake Thibault are the only interconnected lakes in the study area, with Thibault located upstream. Each lake involved in this study sustain relatively simple food webs, where only *Salvelinus* species can be found. The *Salvelinus* species are Arctic char (*Salvelinus alpinus*), brook trout (*Salvelinus fontinalis*), and lake trout (*Salvelinus namaycush*).

Table 1. Lake characteristics of the four study lakes in Gaspésie National Park.

Lakes	Altitude m.a.s.l.	Surface area (ha)	Deepest point (m)	Specie composition
Cascapédia	500	129	18.3	Arctic char, brook trout
Haymard	683	8	3.0	brook trout
Paul	469	74	14.0	Arctic char, brook trout, lake trout
Thibault	515	47	15.2	brook trout, lake trout

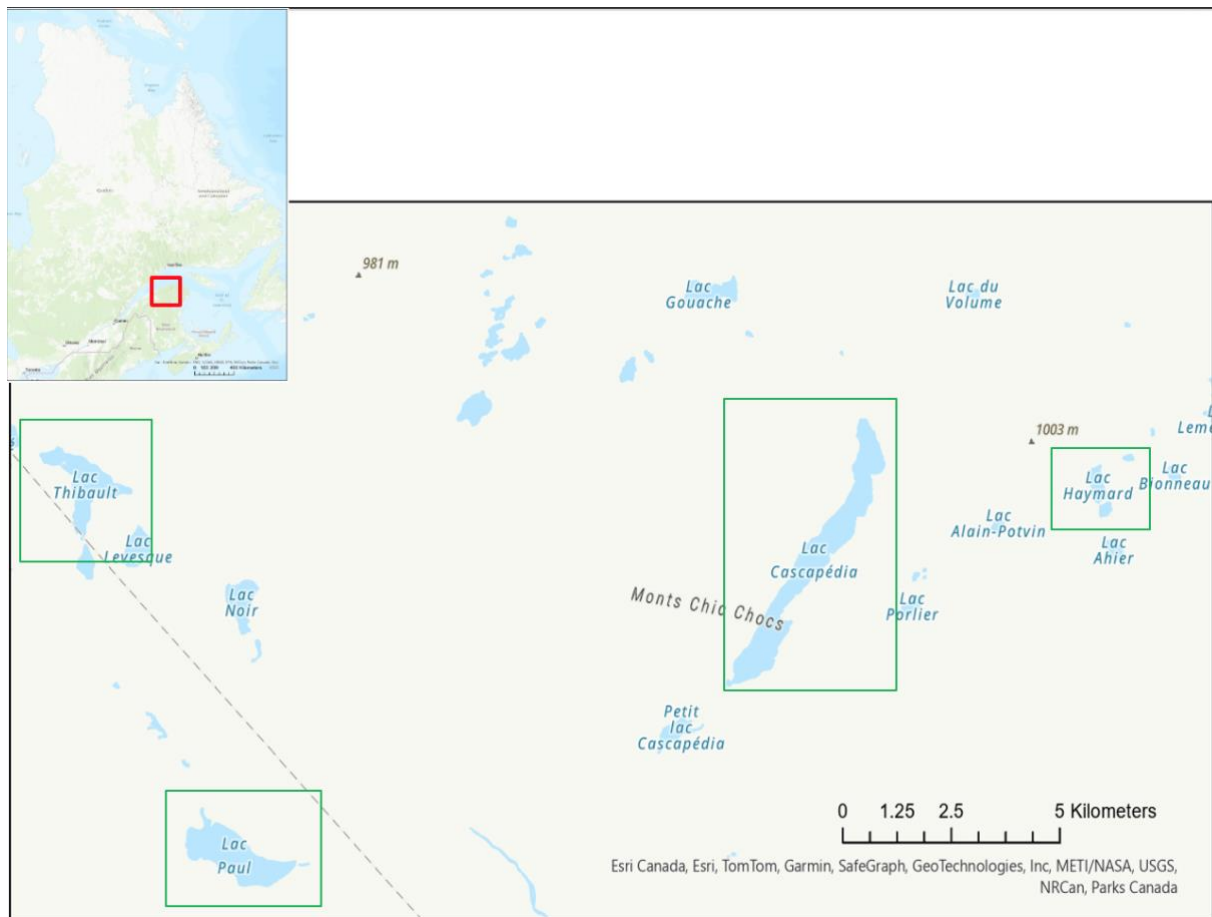


Figure 1. The map shows the location of the study area with the selected lakes in Gaspésie National Park in Québec (Canada; $\sim 48^{\circ}56'N$, $66^{\circ}14'W$). The upper left corner shows a map of the region of Québec where the study area is marked with the red square. All lakes where brook trout were captured are marked with green squares.

2.2. Field sampling

During the winter season, brook trout were captured in three of the study lakes (Cascapédia, Paul, and Thibault) by using a variation of different equipments, including multimesh sinking gillnets, lobster traps, and tip-ups. The gillnets were typically a standard Nordic net with the dimension of 30m in length and 1.5m in depth. These gillnets have a total of 12 different mesh sizes ranging from 5-55mm, where each section is 2.5m long. The goal was to sample a maximum of 50 individuals from each lake, which resulted in a total number of 135 individuals. All winter fieldwork took place in the period between 15th of March to 30th of March 2023.

Summer fieldwork was conducted in the period between 5th of July to 31st of July 2023, where the goal was also to sample a maximum of 50 individuals per lake, resulting in a total of 195

individuals. Multimesh sinking gillnets was the only sampling equipment used for capturing brook trout during summer. These gillnets were typically North American brook char nets with the following dimensions: 22.8m in length, 1.8m in depth, and 6 different mesh sizes ranging from 25-76mm, with each section being 3.8m long. All sampling equipment's used to capture brook trout were conformed to the standardization guidelines for ichthyological sampling in inland waters established by the Ministry of Forests, Wildlife, and Parks (MFFP). All nets deployed were placed in predetermined strata, close to the lake bottom, and perpendicular to the shore. GPS position of each net was recorded (*Figure 2*), along with the depth of both start and end position of the net. The nets were left in the water for ~24 hours before retrieval. For both seasons, the following measurements were taken from each individual: total length (mm), body weight (g), sex, maturity, and gonad weight (g). The gut contents together with all other digestive organs were collected, frozen, and transported back to Norway for laboratory work.

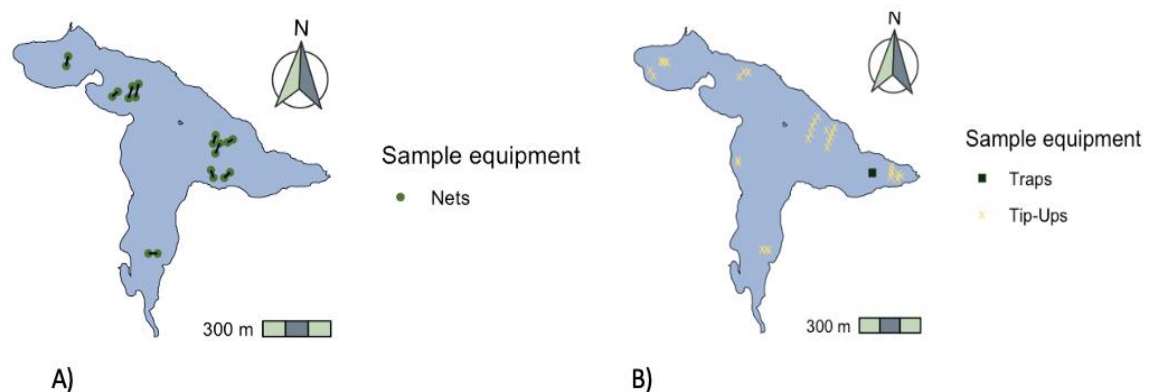


Figure 2. The map is showing sample equipment locations where brook trout was captured during summer (A) and winter (B) of 2023 for Lake Thibault. Sampling equipment's are color coded and with different shapes. See Fig. AX for similar maps of the other selected lakes.

2.3. Laboratory work

Laboratory work was conducted at the Norwegian University of Life Sciences from October 2nd to December 18th, 2023. A total of 330 individuals underwent stomach content analysis, with the distribution across lakes as follows: Cascapédia; 50 summer + 52 winter, Haynard; 47 summer only, Paul; 55 summer + 37 winter, and Thibault; 45 summer + 44 winter. The sampling of stomach content involved categorizing prey items to the taxonomic level of 'order' for all invertebrates with the help of a stereomicroscope. All invertebrates that was not

possible to identify to a taxonomic order was instead recorded as 'unknown invertebrate'. Prey fish was identified to the taxonomic level of 'specie' unless digestion obscured identification, in which case, they were recorded as 'fish unknown'. All prey items, regarding both fish and invertebrates, that were below 50% of the original body mass were recorded as a partial prey item for its specific prey category. For a few stomachs that contained a large number of partial prey items of zooplankton or macroinvertebrates, a subset was made to estimate prey number and prey mass. The subset was utilized by extracting all whole prey items from the sample leaving only partial prey items. Then, the weight of the first 100 partial prey items was used to determine the number of prey items left in the sample by weighting them. Subsets was only necessary in Lake Paul, where stomachs often contained hundreds of smaller prey items such as mayfly larvae (*Ephemeroptera*) and *Diptera* larvae. Additionally, all items that were not possible to put in a specific prey category was counted as miscellaneous remains. All prey items were weighted (M) and counted (N).

2.4. Relative importance index

First, the relative importance index (RI) was used to represent prey items from stomach contents (George & Hadley, 1979). All stomachs containing prey items was selected with the exception of stomachs only containing miscellaneous remains, parasites and empty stomachs. The number of individual stomachs used to calculate RI was reduced from 330 to 243 individuals with the distribution across lakes as follows: Cascapédia; 32 summer + 38 winter, Haymard; 35 summer, Paul; 46 summer + 31 winter, Thibault; 34 summer + 27 winter. To examine the RI for each prey item in a population based on seasonal and temporal variation, RI was calculated by using individuals from each lake and season independently, resulting in a total of 7 different calculations. The same procedure was repeated two times over for prey groups including fish vs. zooplankton vs. benthic macroinvertebrates vs. terrestrial prey items, and between fish vs. invertebrates. In a given fish population (lake), the relative importance of prey taxon i is determined by the following equation:

$$RI = AI \times 100 / \sum_i^n AI$$

1)

where $AI (= \%O + \%N + \%M)$ is the absolute importance of prey taxon i within a population, where $\%O$ denotes the percentage of all non-empty stomachs containing prey taxon i , $\%N$

represents the percentage proportion of prey taxon i items in relation to the total number of prey items in all stomachs, %M indicates the percentage proportion of prey taxon i in mass relative to the total mass of prey items in all stomachs, and n signifies the number of prey taxa (Stevens et al., 1982; Hulsman et al., 2016)

2.5. Stomach content analysis

2.5.1. Diet overlap and individual specialization

All data analysis was conducted in R (version 4.3.3, 2024-02-29). To assess the degree of individual specialization among individuals of different populations and between season, indices related to individual specialization were examined. All data were filtered with the *dplyr* package (Wickham et al., 2023) and were based on count data N of the specific prey items and analyzed using a R package '*R Individual Specialization*' (*RInSp*) (Zaccarelli, 2013). Diet overlaps were determined by the 'Within Individual Component/Total Niche Variation' (WIC/TNW), which takes the variation of prey consumed by the individuals in a population divided by the overall variation of prey items in the same population (Roughgarden, 1974). The values generated are ranging from 0 to 1 and are assessing the individual degree of overlap relative to the total amount of consumed prey items in the same population. As the value approaches 1, individuals utilize more of the population's niche width giving an increased inter-individual overlap, meaning they are showing generalistic feeding behavior, whereas lower values indicate decreased inter-individual overlap, hence higher grade of specialization (Bolnick, 2002; Araújo et al., 2008).

E index was assessed to measure the average pairwise dissimilarity between individuals related to evenness of prey items in a population. Therefore, an estimation of inter-individual niche variation was obtained, where values close to 0 indicate high inter-individual niche variation, meaning that individuals are more specialized because the distribution of prey items is unevenly spread within a population. Conversely, values close to 1 represent low inter-individual niche variation and indicate higher generalistic feeding behavior because prey items are more evenly spread in the population (Zaccarelli, 2013). Additionally, a jackknife estimation of the variance of E is derived based on the estimation procedure provided by Araújo et al. (2008). The index type of the variance of E is based on the Saramäki index which reduce the influence from small sample sizes (Saramäki et al., 2007). Additionally, the relative degree of clustering in a network was tested to examine modularity in the niche

variation of discrete groups in a population, measured as C_{wi} . (Saramäki et al., 2007; Araújo et al., 2008) as follow:

$$C_{wi} = \frac{1}{k_i(k_i - 1)} \sum_{j,h} (w_{ij}w_{ih}w_{jh})^{\frac{1}{3}} \quad 2)$$

Where k_i is the number of edges between individual i and its neighbors. w_{ij} is the weight of the edge between individual i and j obtained by dividing the actual weight by the maximum of all weights. The summation quantifies the weight of all edges between individual i and its neighbors to each other (Zaccarelli, 2013). Values generated from are ranging between -1 to 1. When $C_{wi} =$ zero, it indicates no modularity, meaning that the population is not organized into clusters, but when $C_{wi} > 0$, individuals form discrete groups specializing on distinct sets of resources, hence higher modularity. When the value of $C_{wi} < 0$, the network degree of clustering is lower than what would be expected from the overall network density of connections, meaning that diet variation takes place at the individual level, and not between discrete groups (Zaccarelli, 2013).

Pairwise similarity or dissimilarity between individuals in a population was calculated from a measure of individual specialization based on the average pairwise overlap of niche distribution. It was estimated by using Czekanowski's proportional similarity index (PS_i), which is given by the following formula:

$$PS_i = 1 - \frac{1}{2} \sum_j |p_{ij} - q_j| \quad 3)$$

where p_{ij} is the frequency of prey items j in the individual i 's diet, and q_j is the frequency of prey item j in the population as a whole. Values generated is also ranging from 0 to 1, where 0 indicate complete dissimilarity, meaning that two individuals have completely different prey preference, whereas 1 indicate complete similarity, meaning that two individuals have 100% identical prey preference. Results from the PS_i is then used to determine the degree of specialization (IS) by using the following equation:

$$IS = \frac{\sum_i(PS_i)}{N}$$

4)

The value of IS is most consistent with the true overlap (Linton et al., 1981) calculated by the average of all PS_i values. Values higher than 0.6 often representing significant biological diet overlap, hence a significant generalist feeding behavior within a population (Schoener, 1968; Zaccarelli, 2013).

2.5.2. Diversity index

Diversity among populations was analyzed by using the famous diversity indices of Shannon and Simpson diversity indexes executed by using the R package *vegan* (Oksanen et al., 2022). The two indices work differently where the Shannon diversity index is emphasizing the richness component and respond more to changes in importance of rare prey items, whereas the Simpson diversity index is emphasizing the evenness component and respond to changes of the more dominant prey items within a population (Nagendra, 2002). The inputs in this analysis are the abundances of each prey items, and the indexes is a measure of diversity which considers the abundance and occurrence of different prey items present in individuals of a specific population (Gorelick, 2006). For Shannon diversity index, values are ranging from 0 to infinite, where zero indicate an absence of diversity, and infinite indicate large diversity based on the number of prey items represented in the entire population (Nagendra, 2002). The Shannon diversity index is defined as:

$$SHDI = 1 - \sum_{i=1}^n p_i \times \ln p_i$$

5)

where n is representing the number of prey items and p_i is the relative abundance (probability) of prey item i , estimating which prey item a randomly selected sub-unit of the population will belong to (Nagendra, 2002; He & Hu, 2005; Gorelick, 2006).

The Simpson index is defined as:

$$SIDI = 1 - \sum_{i=1}^n p_i^2$$

6)

estimating the probability that individuals within a population have equally abundant diversity of prey items in the stomach content. The index ranges from 0 to 1, where 0

indicates no diversity, meaning that only one prey item is present, whereas 1 indicates infinite diversity, meaning all prey items are equally abundant (Nagendra, 2002).

2.5.3. Generalized Linear Models (GLM)

Generalized linear models were used to examine presence-absence relationship in feeding within a fish population by utilizing the R package 'glm2' (Marchner 2011). All stomachs were categorized into two groups: empty versus non-empty stomachs, which was given the values 1 and 0, respectively. Models focusing solely on seasons were developed by segregating individuals from each lake, with the exception of Lake Haymard, which only contained data from the summer season, and then use a GLM to examine the importance of seasonal variation for each lake independently. Moreover, the 'lme4' package (Bates et al. 2015), were used to create a generalized linear mixed model where lakes serve as random variables and seasons serve as fixed variables by using the function `glemer.nb`.

The relative importance of the various prey groups (fish, zooplankton, benthic- and terrestrial invertebrates) was used in another mixing model to investigate if lake, season, and other fish physiological variables (total length, sex, maturity, and condition factor) would affect the relative importance of the different prey groups on an individual level. Two R packages, 'mvabund' and 'glmmTMB' (Brooks et al. 2017; Wang et al. 2022) were used, and all variables was treated as fixed variables, while individual Fish ID was used as a random variable. To identify the best model, variables with the highest p-value were eliminated from the mixing model (GLMM) until the model retained the variable with lowest p-value, which make the best suited model for the analysis. The best suited model was analysed by using the function `'anova.manyglm'` in R.

The mean and standard deviation of total length (mm) and condition factor were calculated to examine the differences in population characteristics to gain knowledge about variables that may alter the results of the stomach content analysis. A statistical analysis with ANOVA was conducted to check for statistical significant differences for total length between populations across lake and season.

3. Results

3.1. Relative importance

A total of 243 individual stomachs provided the basis for calculating the Relative Importance (RI) across all four study lakes and for both seasons, revealing a total of 25 distinct prey items spanning various taxa (*Figure 3*). The combination of consumed prey items and their relative importance demonstrated considerable variability, attributable to spatial and temporal changes. Despite this variability, the dietary pattern remained generally consistent across lakes, where prey items such as *Ephemeroptera*, *Odonata*, *Zygoptera*, or *Trichoptera* being highly dominant depending on lake and season.

In Lake Paul, *Ephemeroptera* constituted just over 60% of the stomach content during summer and ~40% during winter. Conversely, in lakes such as Cascapédia and Haymard, *Odonata* emerged as the most dominant prey item, contributing over 30% to stomach content. While both *Ephemeroptera* and *Odonata* made substantial contributions during both seasons, prey items like *Zygoptera* and *Trichoptera* exhibited a notably larger presence in stomach content during winter (10-30%) compared to summer (<1%). It's noteworthy that all these prey items are classified as benthic macroinvertebrates, initially believed to constitute the primary contribution to stomach content.

The majority of fish sampled from stomach contents were classified as 'fish unknown' due to digestion obscuring identification. However, one fish species ('SAFO') was identified in stomach contents from Lake Haymard, where brook trout is the sole fish species present, rendering further identification unnecessary. Generally, fish were infrequently detected in stomach content and were only observed in Lake Cascapédia and Lake Haymard.

Consequently, fish played a minor role in stomach content contribution, accounting for only three to six percent of the RI depending on lake and season (*Figure 4*).

Prey items designated as 'ZooplanktonParts' emerged as the most consumed prey item from the zooplankton category (RI = 20% in Thibault, 16% in Haymard, and 12% in Cascapédia during summer), except in Lake Paul, where zooplankton were generally underrepresented during both seasons (*Figure 3*, *Figure 4*). Other prey items from the zooplankton taxa with the highest RI value after 'ZooplanktonParts', were *Cladocera*, followed by *Bosmina* and *Copepoda*, which was only found in a few stomachs contributing minimally to the overall

stomach content. Interestingly, *Copepoda* was only detected during winter and in Lake Cascapédia (Figure 3).

Terrestrial prey items, logically only recorded during summer season, comprised a group of prey items with varying contributions to stomach content, with *Coleoptera* prey items being the most prevalent in this prey item group (Figure 3). *Coleoptera*, designated as 'ColeopteraTerr', revealed an interestingly 20% contribution to stomach content in Lake Thibault, in contrast to Lake Paul where it was relatively absent. Remarkably, Lake Cascapédia was the only lake where all four prey items in the terrestrial prey item group were obtained from stomach content, consisting of 'ColeopteraTerr', 'OdanataAdult', 'Wasp', and 'Formicidae'.

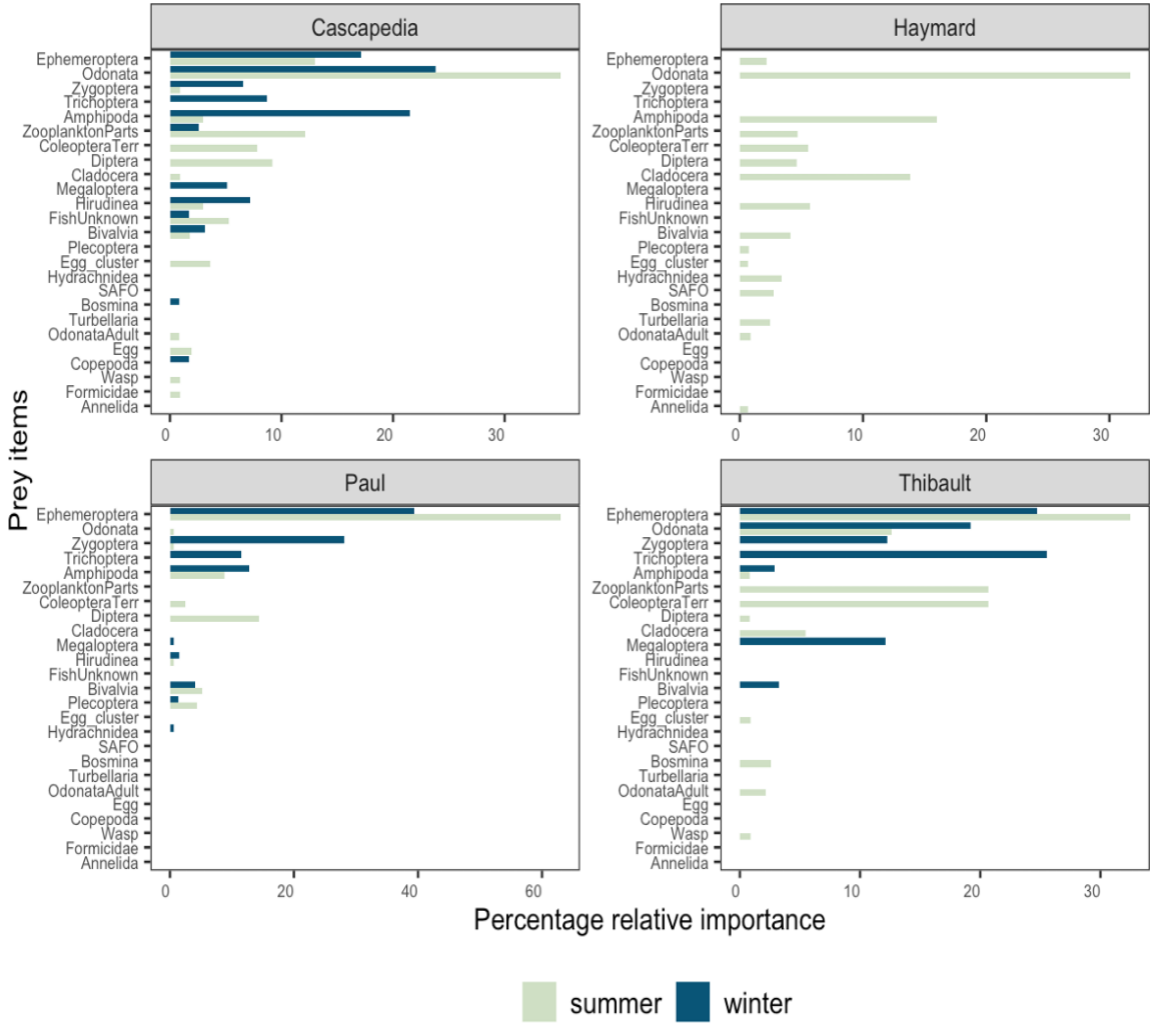


Figure 3. Relative importance (RI) for all 25 different prey taxa extracted from the stomach content in the period between October 2nd to December 18th, 2023, including all four study lakes in Gaspésie National Park and the seasonal variation as color coded.

Across all four lakes and throughout the seasons, benthic macroinvertebrates consistently emerged as the predominant prey group in the diet of brook trout populations (*Figure 4*). However, their contribution varied significantly among the lakes. In Lake Paul, benthic macroinvertebrates stood out, constituting nearly 100% of the total diet composition during both seasons, illustrating their heightened importance. In contrast, their contribution to the overall diet was notably lower in populations found in Lake Cascapédia, Haymard and Thibault during summer, comprising ~50-65% of the diet composition. Additionally, the contribution of benthic macroinvertebrates to stomach content exhibited seasonal variation in Lake Cascapédia and Lake Thibault. The data suggest that they are more important contributors to stomach content during the winter season, reaching ~95% and 100% contribution in Lake Cascapédia and Lake Thibault, respectively.

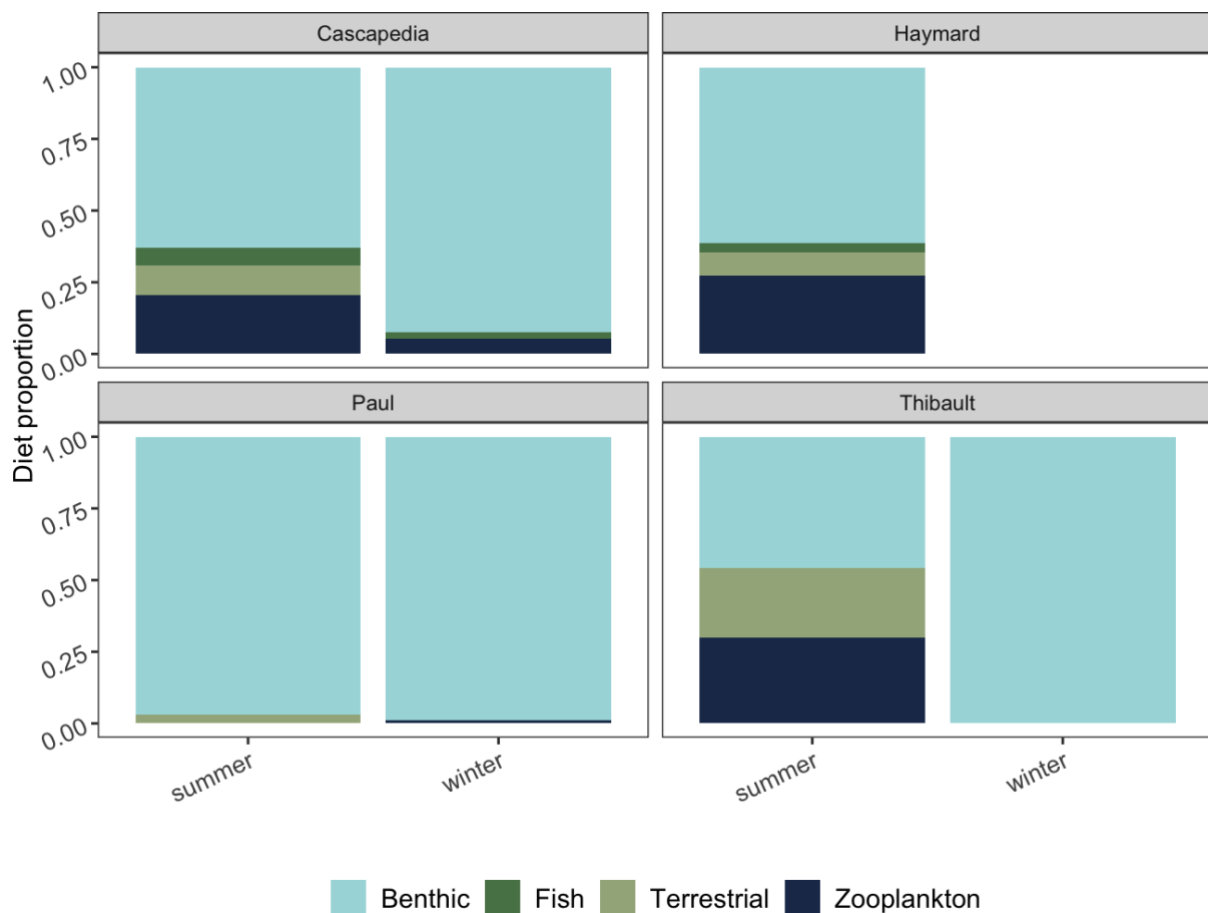


Figure 4. Overview of the different prey groups found in the stomach content for brook trout in the lakes of Gaspésie National Park. The figure shows the total consumption rate for each of the four prey groups (benthic- and terrestrial invertebrates, zooplankton, and fish) and their overall contribution to stomach content for both lake and season.

Conversely, the contribution of other prey groups (zooplankton, terrestrial invertebrates, and fishes) varied depending on the lake and season. For instance, in Lake Thibault, there was a marked shift in prey consumption from summer to winter. During summer, both terrestrial prey items and zooplankton contributed ~25% each to the total diet composition, whereas none of these prey groups are found in stomach content during winter. A similar trend, though less pronounced, was observed among the brook trout population in Lake Cascapédia, where contributions from zooplankton, terrestrial invertebrates and fishes in stomach content decreased from ~35% to ~5% contribution from summer to winter. (Figure 4).

3.2. Dietary analysis

The dietary analysis conducted across the study lakes revealed a diverse range of feeding strategies among the fish populations within Gaspésie National Park. Various metrics including overlap, Araújo's *E index*, individual specialization (derived from the average measurement of PS_i), and network clustering (C_{wi}) were measured, as summarized in Table 2.

Table 2. Values from the overlap and individual specialization measurements (WIC/TNW, Araújo's E value, IS, and C_{wi}) indices for all four study lakes in Gaspésie National Park.

Lake	Season	WIC/TNW (Overlap)	E value	IS	C_{wi}
Cascapédia	Summer	0.53	0.83	0.20	0.31
Cascapédia	Winter	0.60	0.81	0.27	0.32
Haymard	Summer	0.70	0.79	0.14	0.10
Paul	Summer	0.50	0.46	0.62	0.05
Paul	Winter	0.64	0.56	0.59	-0.04
Thibault	Summer	0.47	0.72	0.10	0.13
Thibault	Winter	0.27	0.78	0.30	0.31

Overlap values, calculated from Within Individual Component / Total Niche Variation (WIC/TNW), highlighted distinct feeding patterns among brook trout populations. Notably, Lake Thibault exhibited the lowest inter-individual overlap, indicating higher degree of specialization (Figure 5). Remarkably, this population also displayed significant seasonal variation, with overlap values of 0.47 and 0.27 for summer and winter, respectively, a variation not observed in other lakes. During summer, populations in Lake Cascapédia, Paul

and Thibault demonstrated similar overlap levels (0.50, 0.53, and 0.47 respectively), suggesting that the populations do not generalize or specialize on specific prey items. However, Lake Haymard stood out with an overlap value of 0.70, indicating a more generalistic feeding strategy among that population. Furthermore, during winter, overlap increased in populations in Lake Cascapédia and Paul (0.60 and 0.64, respectively), whereas Lake Thibault maintained lower overlap, signifying significant specialization (0.27).

Araújo's *E index*, measuring inter-individual niche variation, generally exceeded 0.70 for most populations, except for Lake Paul maintaining values of 0.46 and 0.56 for summer and winter, respectively (*Table 2, Figure 5*). From the same calculation, the degree of clustering in a network was obtained, revealing different degree of clustering among lakes. Lake Cascapédia exhibited minimal seasonal variation (0.31 versus 0.32) yet maintained the highest degree of network clustering. However, the establishment of discrete specialized groups within populations was not significant. Interestingly, Lake Paul displayed negative degree of clustering during winter (-0.04), suggesting that the diet composition was more influenced by individual preference rather than preferences of a discrete groups. The lake with the highest seasonal variation was Lake Thibault (0.13 during summer and 0.31 during winter).

Furthermore, *Figure 5* illustrates a higher degree of individual overlap in Lake Paul, as indicated by Czekanowski's proportional similarity index (PS_i) (0.62 and 0.59 for summer and winter, respectively). This result indicate that two individuals are more likely to have the same diet in Lake Paul compared to the other lakes, and these results aligned with the RI values, where *Ephemeroptera* having more than 60% relative importance to stomach content in Lake Paul. Additionally, the *IS* based on pairwise dissimilarity or similarity is indicating that pairwise individuals in the populations in Lake Cascapédia, Haymard and Thibault is significantly specialized in their feeding strategy which does not align with the results form diet overlap measured from WIC/TNW.

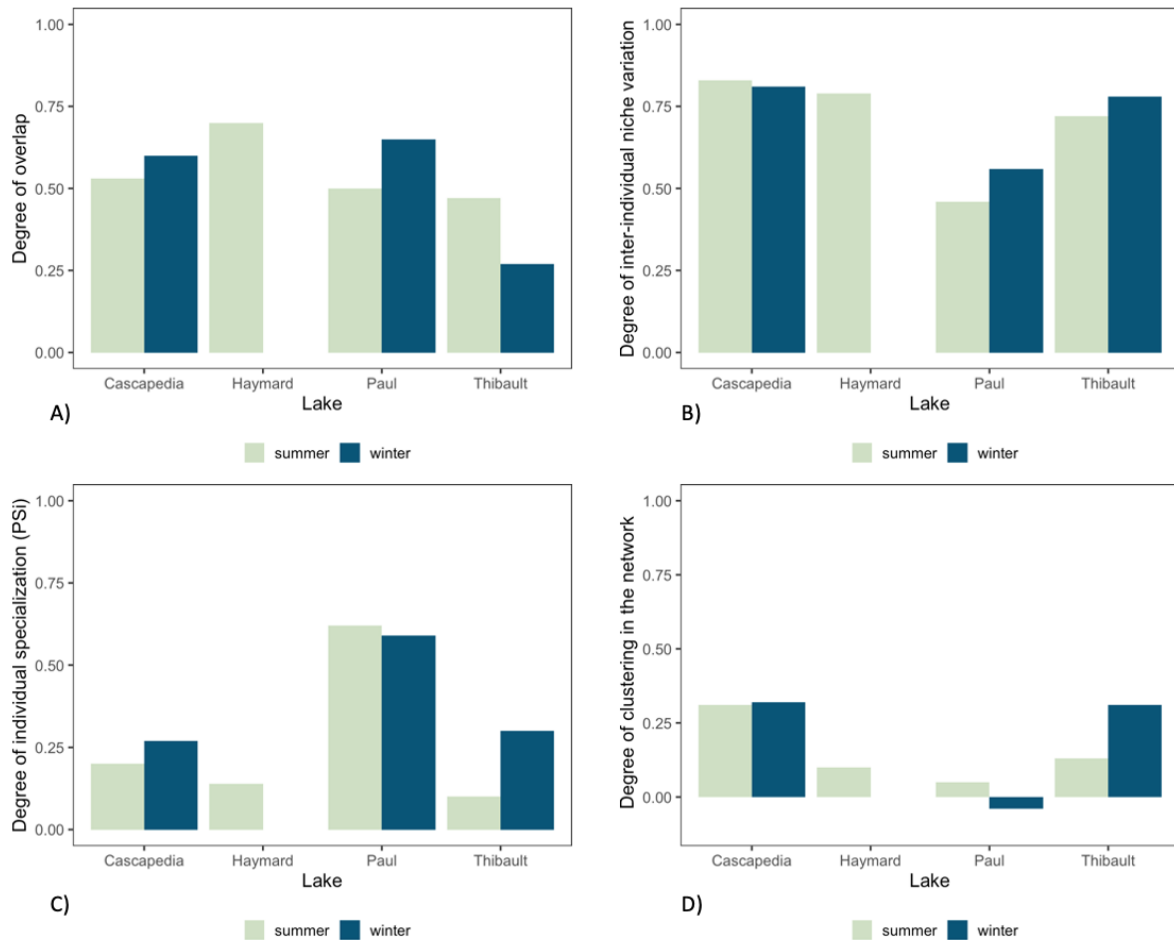


Figure 5. Dietary analysis were examined on population level by measuring A) individual overlap, B) inter-individual niche variation based on Araújo's E index, C) individual specialization based on similarity/dissimilarity index of the PSi, and D) degree of clustering in a network.

Results from the Shannon and Simpson diversity indices show that populations obtain various diversity of prey items in the stomach content depending on lake and season (Figure 6). The Shannon indices reveal that Lake Paul harbors the greatest diversity, influenced by both richness and abundance of rare prey items, with values of 0.46 and 0.51 during summer and winter, respectively. Similarly, the population in Lake Thibault is also exhibits high diversity during summer (0.45), but strongly declines during winter (0.10), indicating significant seasonal variation. Apart from these instances, brook trout populations generally demonstrate low diversity based on the Shannon diversity indices (Table 3, Figure 6).

In terms of the Simpson indices, both summer and winter seasons in Lake Paul had the highest values of 0.28 and 0.27, respectively, together with summer season in Lake Thibault (0.25). Notably, Lake Thibault also displayed the highest seasonal variation, with values of

0.25 during summer and 0.04 during winter. Overall, these values suggest high diversity based on evenness, as low values indicate that prey items were relatively equally abundant. Consequently, the results revealed that individuals tend to exhibit generalistic feeding behavior.

Table 3. Values from the Shannon and Simpson indices for all four study lakes in Gaspésie National Park.

Lake	Season	Shannon diversity	Simpson diversity
Cascapédia	Summer	0.16	0.08
Cascapédia	Winter	0.22	0.10
Haymard	Summer	0.26	0.15
Paul	Summer	0.46	0.28
Paul	Winter	0.51	0.27
Thibault	Summer	0.45	0.25
Thibault	Winter	0.10	0.04

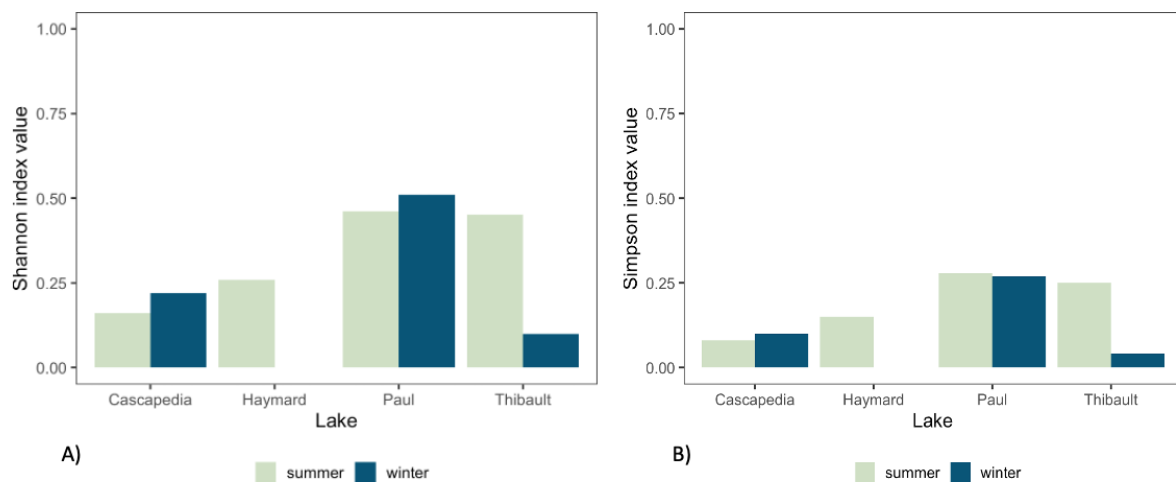


Figure 6. Diversity of prey items in stomach content were examined on population level by using A) the Shannon indices and B) the Simpson indices.

3.3. GLM and GLMM

The mean and standard deviation of total length and condition factor highlights differences in these variables. Generally, the condition factor is lower during winter, suggesting a higher metabolic rate relative to consumption rate (*Table 4*). These findings align with the observation of a higher percentage of empty stomachs during winter compared to summer, indicating reduced foraging behavior. The mean total length of the populations ranges between 191mm and 332mm (*Table 4, Figure 7*). However, statistical analysis revealed that there was no statistical length difference among the populations of Gaspésie National Park ($p = 0.473$).

Table 4. Population characteristics based on the mean and standard deviation of total length (mm) and condition factor, and Percentage empty stomachs.

Lake	Season	Length total/LT (mm) Mean \pm SD	Condition factor Mean \pm SD	Percentage empty stomachs
Cascapédia	Summer	191.49 \pm 56.18	1.25 \pm 0.16	9.62
Cascapédia	Winter	237.33 \pm 58.00	1.13 \pm 0.20	13.46
Haymard	Summer	261.89 \pm 99.09	1.33 \pm 0.13	20.83
Paul	Summer	217.86 \pm 57.46	1.25 \pm 0.24	3.57
Paul	Winter	269.68 \pm 82.49	1.09 \pm 0.40	5.41
Thibault	Summer	262.20 \pm 80.83	1.14 \pm 0.24	6.67
Thibault	Winter	332.81 \pm 61.81	1.00 \pm 0.09	11.36

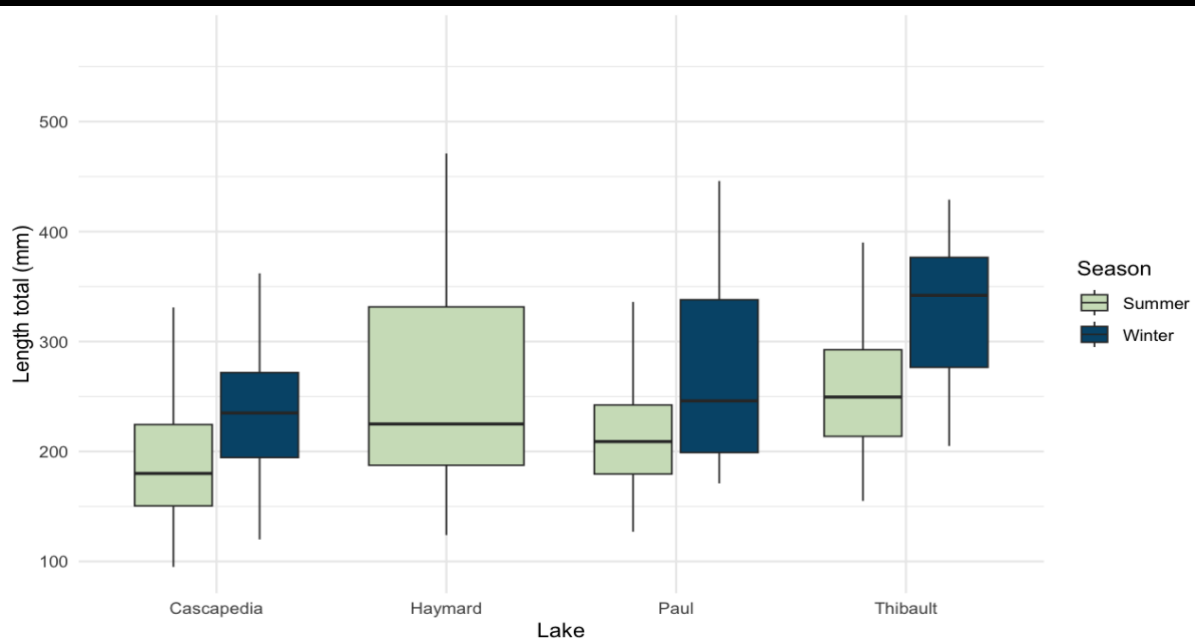


Figure 7. the distribution of brook trout total length for all lakes and seasons as color coded.

Generalized Linear Models (GLM) examining the relationship between empty versus non-empty stomachs, based on season for each lake independently, yielded non-statistically significant p-values: Cascapédia = 0.495, Paul = 0.985, and Thibault = 0.069. Additionally, the Generalized Linear Mixing Model (GLMM) utilizing lakes as random variables and seasons as fixed variables for the entire population (330 individuals) also gave statistically non-significant values ($p = 0.633$) when tested for empty versus non-empty stomachs. These findings suggest no statistically significant difference between the occurrence of starving versus foraging fish based on temporal and seasonal changes.

Table 5. The mixing model eliminates each variable by taking out the variable with the highest p-value from each model, ending up with model best suited for this research (GLMM6).

Model	Season	LT	Lake	Condition factor	Sex	Maturity
GLMM1	0.035	0.062	0.044	0.148	0.902	0.984
GLMM2	0.017	0.060	0.078	0.498	0.764	-
GLMM3	0.013	0.043	0.055	0.495	-	-
GLMM4	0.026	0.041	0.075	-	-	-
GLMM5	0.003	0.186	-	-	-	-
GLMM6	0.003	-	-	-	-	-

The results of the ANOVA analysis, conducted by using the best suited model for this research (GLMM6), provided valuable insights into prey group preference. Utilizing the `anova.manyglm` function on the GLMM6 model revealed statistically significant values ($p = 0.001$) for seasonal variability, indicating that season played a vital role in determining which prey group an individual brook trout choosed to feed on.

4. Discussion

4.1 Relative Importance Index

The study revealed significant variations in the diet composition of brook trout across different seasons and lakes within the Gaspésie National Park. After examining the Relative Importance of prey items and prey groups in the different study lakes, the data demonstrated that brook trout predominantly feed on benthic macroinvertebrates in the littoral zone, with notable contributions from zooplankton and terrestrial prey items during summer in Lake Cascapédia and Lake Thibault. These results align with the results from a study conducted by Tiberti et al., (2016). Additionally, RI for benthic macroinvertebrates went from 50% contribution to stomach content during the summer season, to 100% contribution during the winter season. Such drastic changes might indicate a switch in prey availability, at least for terrestrial prey items because they are not present during winter season. These findings align with previous studies suggesting that feeding behavior among brook trout populations, are able to be altered based on the availability of prey items (Jirka & Kraft, 2017). However, the relative importance of prey items varied among lakes, where some lakes exhibiting higher contributions for specific prey items such as *Ephemeroptera* or *Odonata*. It also revealed large differences of contributing prey items depending on seasonal variation, where *Trichoptera* and *Zygoptera* had much higher contribution to stomach content during winter season, especially in Lake Paul and Lake Thibault. All these variations could be attributed to differences in habitat characteristics and prey availability among lakes (Browne & Rasmussen, 2009). A know principal in ecology say that larger habitats often obtain more species because they are capable of housing more niches and different niches. This principal aligns with the examination of stomach content in this study where a total of 20 different prey items was found in Lake Cascapédia considering both seasons, contrasting 16 prey items in Lake Haymard, 15 prey items in Lake Thibault, and 12 prey items in Lake Paul.

4.2 Stomach content analysis

The results of stomach contents analysis revealed many interesting aspects related to diet overlaps, prey diversity and prey abundance in the different populations of Gaspésie National Park. The analysis showed that individuals within a population had rather similar diets indicating individual overlap in Lake Haymard (0.70) and to some degree Lake Paul during winter (0.64). Individuals within the populations in Lake Cascapédia during both seasons, and

in Lake Paul and Thibault during summer season, expressed no significant overlap, or significant specialization. Only Lake Thibault during winter season displayed significant less diet overlap between individuals, indicating individual specialization. One possible explanation for this might be because of prey availability or prey abundance. Winter season creates ice cover that block prey items from entering into the lake making it less of a choice to specialize on benthic macroinvertebrates instead. Another possibility is that cold water favor foraging behavior for lake trout in Lake Thibault, and that they start to dominate in the littoral zone forcing brook trout to feed on less optimal prey. In this case that would be *Trichoptera* larvae because fish often get stomachs full of wood when eating *Trichoptera* larvae. Moreover, brook trout used a more diverse set of prey items depending on lake and season, relying much more on zooplankton and terrestrial prey items during summer compared to winter in Lake Cascapédia and Lake Thibault. Conversely, in Lake Paul brook trout is more or less solely depended on benthic macroinvertebrates. With the exception of Lake Paul, all lakes displayed a more diverse set of prey items during summer compared to winter, which might not be strange due to terrestrial prey items are not present during winter season. However, all lakes illustrated the importance of benthic macroinvertebrates, and that brook trout highly rely on littoral resources. Although this is not certain, there is a possibility of high interspecific competition in Lake Cascapédia, Paul, Thibault because brook trout live sympatrically with Arctic char and lake trout in these lakes. This is especially a concern during winter season, given the fact that both of Arctic charr and lake trout are more cold-water adapted than brook trout, and they are obtaining very identical niche width (Behnke, 2002). Therefore, during every winter season, Arctic char and lake trout may possibly have a competitive advantage over brook trout. However, populations of several species might be able to live sympatrically, at least for a short while, and feed on the same resource if the resource is very abundant. Despite this, it is clear that fish species with largely overlapping diets, and potentially utilizing the exact same niche, will result in one specie likely exclude the other (Hardin 1960).

Results from the analysis of *Araújo's E index* indicate a consistent pattern across lakes and seasons, revealing that prey items tend to be evenly distributed within the populations and across most lakes (Cascapédia, Haynard, and Thibault). This result can maybe be explained by saying that a few individuals having very full stomachs making an unevenly distribution of the most dominant prey item (*Ephemeroptera*, and *Zygoptera* during winter).

4.3 GLM and GLMM

The study also investigated the relationship between seasonal variation and the occurrence of empty stomach finding no significant results. However, there was trends suggesting that season did have an effect on the number of empty stomachs occurring in a population, especially in lake Thibault where $p = 0.07$.

The study also investigated the influence of physiological traits, such as total length and condition factor to see if these variables were altering the feeding pattern expressed from the results. The result from the stomach content mixing model showed that neither condition factor nor seasonal variations had significant impact on diet preferences. summer and winter. Additionally, the analysis of empty versus non-empty stomachs revealed no statistically significant differences based on temporal and seasonal changes, suggesting that brook trout maintain consistent foraging behavior regardless of environmental conditions.

4.4 Limitations of this study

It is essential to acknowledge the limitations of this study, including potential biases associated with stomach content analysis and the use of sampling techniques. Only using stomach content to analyze diet composition is not something that should be considered and supported techniques, such as Stable Isotope Analysis (SIA), should be added to the methodology. That is because stomach content analysis has a hard time analyzing long-term feeding strategy, but with SIA this issue is solved.

Other limitations to this study include sampling of fish during winter season, where the author did not sample materials for the winter season and could therefore not influence the sampling. First, the habitats of which the fish is captured are very different from each other regarding fish assemblage, surface area and lake depths. This may alter many aspects related to trophic interactions with more or less intra- and interspecific competition, prey availability, and prey abundance.

Interspecific competition is not directly assessed in this study because there are no analysed data about Arctic char or lake trout but are instead representing a theoretical idea that spatial changes are mainly driven by species interaction with the other *Salvelinus* species increasing

competition and prey availability. Future research should focus on the examination of species living sympatrically with brook trout to provide a more comprehensive understanding of trophic interactions, food web dynamics, interspecific competition in alpine lake ecosystems.

5. Conclusion

In conclusion, the results of this study show dietary niche overlap measuring individually, pairwise, and between groups feeding strategy for brook trout species inhabiting alpine lakes in Gaspésie National Park. The results indicate various degree of overlap depending on seasonal and temporal variation but also depending on the stats. Diet overlaps measured from the WIC/TNW indicated that the majority of the populations were expressing neither generalistic or individual specialization except from Lake Haymard (generalistic) and Lake Thibault during winter (Specialist). However, this was challenged by another measure of Individual Specialization IS, where all lakes except Lake Paul exhibited specialist feeding behavior based on the likelihood of two individuals utilizing the exact same niche. Dietary preferences are likely to be largely depended on prey availability, and the dietary patterns in this research show consistency highlighting the importance of benthic macroinvertebrates as primary prey items for each population examined. The observed variations in diet composition, individual specialization, and feeding strategies underscore the complexity of ecological interactions within these ecosystems. By addressing these patterns, this research has contributed to a broader understanding of freshwater ecosystem dynamics in alpine lakes. Additionally, the results from this research can be used to inform conservation efforts, aiming to preserve biodiversity and ecosystem resilience in the face of global environmental changes.

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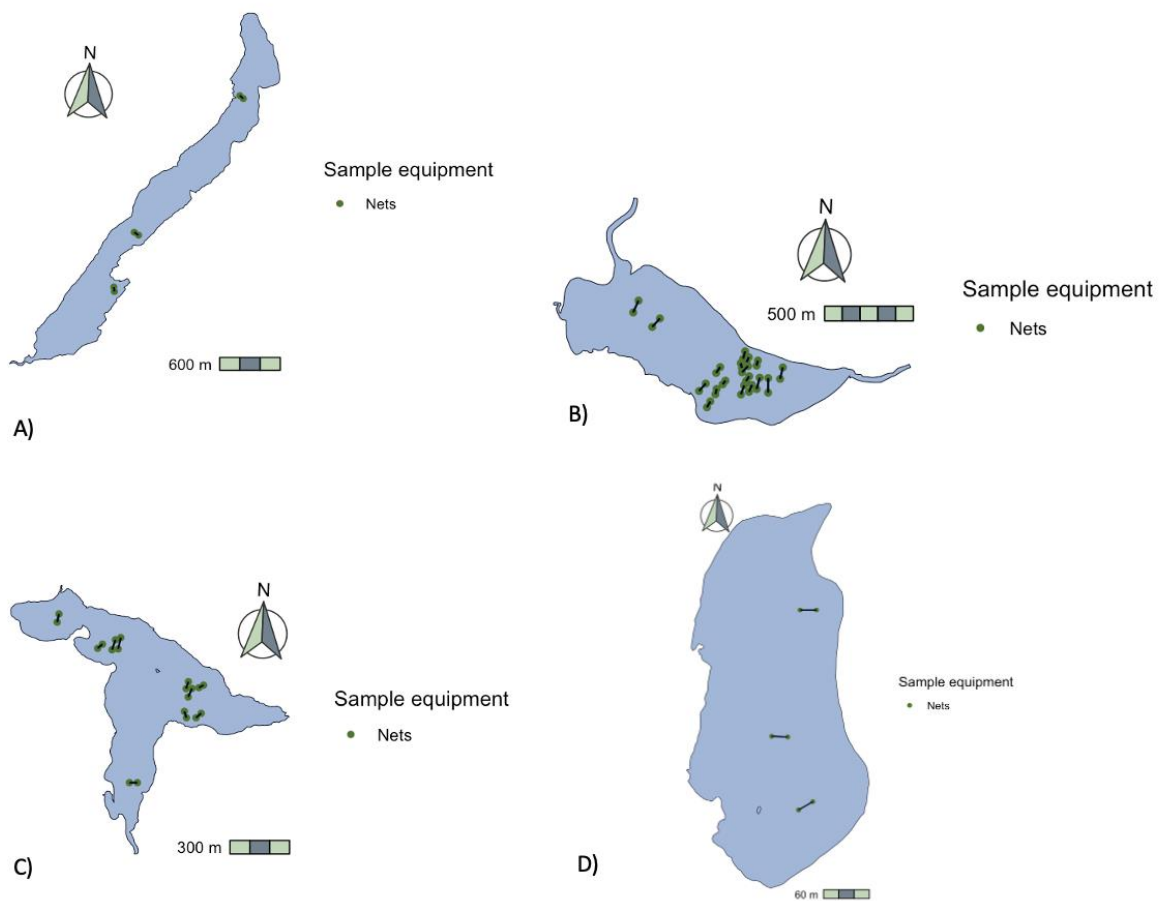


Figure 8. The map is showing sample equipment locations where brook trout was captured during summer of 2023 for all four study lakes – A) Cascapédia, B) Paul, C) Thibault, and D) Haymard. Sampling equipment's are color coded and with different shapes.

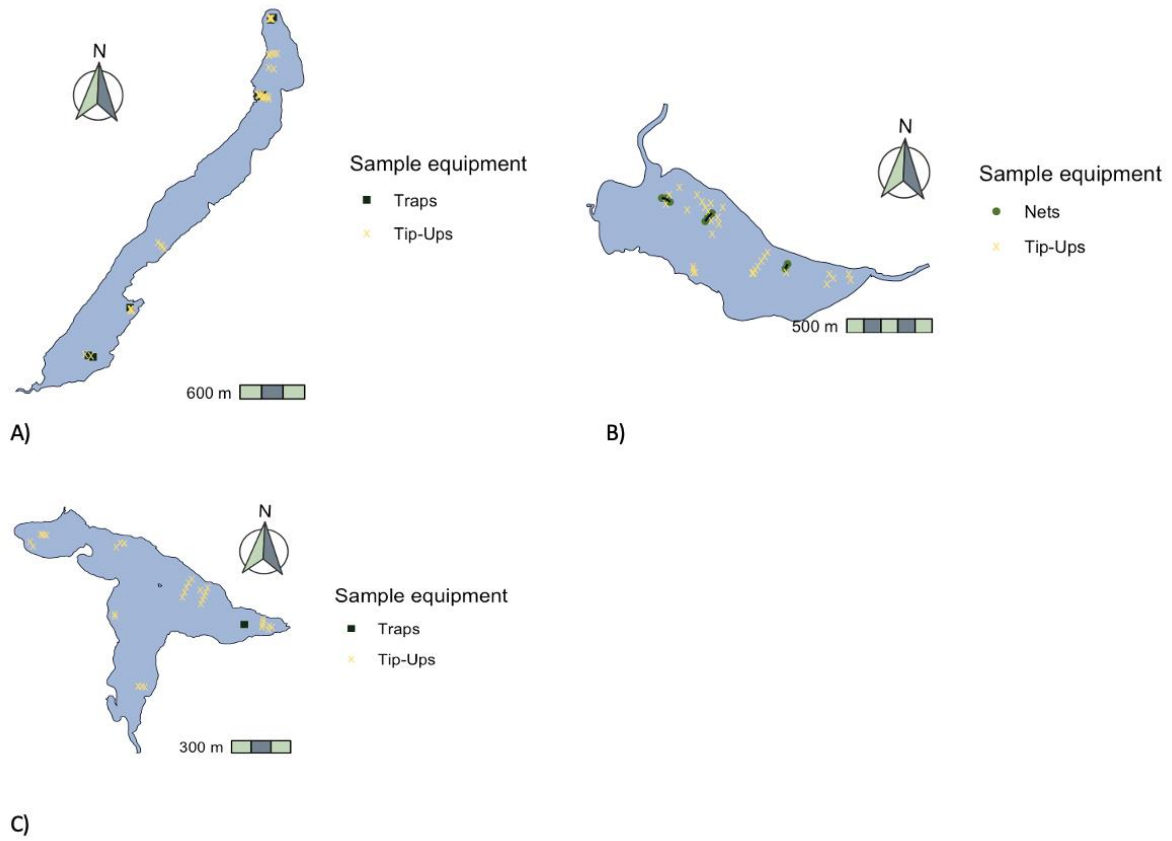


Figure 9. The map is showing sample equipment locations where brook trout was captured during summer of 2023 for all four study lakes – A) Cascapédia, B) Paul, C) Thibault, and D) Haymard. Sampling equipment's are color coded and with different shapes.

R coding

GLM + GLMM

```
library(readxl)
```

```
library(tidyverse)
```

```
#####
```

```
#GLM
```

```
#####
```

```
SAFOMathias <- read_excel("~/Documents/BiologiMaster/Masteroppgave  
/Excel/Excel_files/SAFOMathias (1).xlsx")
```

```
View(SAFOMathias)
```

```
ANOVA_LT <- aov(LT ~ Lake * Season, data = SAFOMathias_1_)
```

```
summary(ANOVA_LT)
```

```
library(ggplot2)
```

```
ggplot(SAFOMathias_1_, aes(x = Lake, y = LT, fill = Season)) +  
  geom_boxplot(size = 0.4, outlier.shape = NA) +  
  labs(x = "Lake", y = "Length total", fill = "Season") +  
  scale_fill_manual(values = c("Summer" = "#cfe0c3", "Winter" = "#005478")) +  
  theme_minimal() +  
  labs(x = "Lake", y = "Length total (mm)")  
  theme(legend.position = "right")
```

```
library(glm2)
```

```
Casc <- filter(SAFOMathias, Lake=="Cascapedia")
```

```
CascGLM <- glm(Casc$Empty_Stomachs~ Casc$Season, family=binomial(link="cloglog"))
```

```
summary(CascGLM)
```

```
Paul <- filter(SAFOMathias, Lake=="Paul")
```

```
PaulGLM <- glm(Paul$Empty_Stomachs~ Paul$Season, family=binomial(link="cloglog"))
```

```
summary(PaulGLM)
```

```

Thib <- filter(SAFOMathias, Lake=="Thibault")
ThibGLM <- glm(Thib$Empty_Stomachs~ Thib$Season, family=binomial(link="cloglog"))
summary(ThibGLM)

#####
#GLM empty stomach
#####

library(lme4)
mod.NB = glmer.nb(Empty_Stomachs~Season+(1|Lake),data=SAFOMathias)
summary(mod.NB)

#####
#GLM negative binomial
#####

library(mvabund)
library(glmmTMB)

GLMMGCol<-SAFOMathias %>%
mutate(RI_Fish=as.numeric(RI_Fish),RI_Benthic=as.numeric(RI_Benthic),RI_Terrestrial=as.
numeric(RI_Terrestrial),RI_Zooplankton=as.numeric(RI_Zooplankton),Season=as.factor(Sea
son),Specie=as.factor(Specie),Lake=as.factor(Lake),Sex=as.factor(Sex),Maturity=as.factor(M
aturity),Condition_Factor=as.numeric(Condition_Factor), LT=as.numeric(LT))

preycounts <- SAFOMathias[, c('RI_Terrestrial', 'RI_Zooplankton', 'RI_Benthic', 'RI_Fish')]
preycounts<-preycounts %>%
mutate(RI_Fish=as.numeric(RI_Fish),RI_Benthic=as.numeric(RI_Benthic),RI_Terrestrial=as.
numeric(RI_Terrestrial),RI_Zooplankton=as.numeric(RI_Zooplankton))

#Convert prey counts to an mvabund object
preymvabund <- mvabund(preycounts)

GLMM1 <- manyglm(preymvabund ~ LT + Sex + Maturity + Season + Lake +
Condition_Factor +(1|IDModel), data = GLMMGCol, family = "negative.binomial")

```

```
drop1(GLMM1, test = "Chisq")
```

```
GLMM2 <- manyglm(preymvabund ~ LT + Sex + Condition_Factor + Season + Lake  
+(1|IDModel), data = GLMMGCol, family = "negative.binomial")
```

```
drop1(GLMM2, test = "Chisq")
```

```
GLMM3 <- manyglm(preymvabund ~ LT + Lake + Condition_Factor + Season  
+(1|IDModel), data = GLMMGCol, family = "negative.binomial")
```

```
drop1(GLMM3, test = "Chisq")
```

```
GLMM4 <- manyglm(preymvabund ~ LT + Lake + Season +(1|IDModel), data =  
GLMMGCol, family = "negative.binomial")
```

```
drop1(GLMM4, test = "Chisq")
```

```
GLMM5 <- manyglm(preymvabund ~ LT + Season +(1|IDModel), data = GLMMGCol,  
family = "negative.binomial")
```

```
drop1(GLMM5, test = "Chisq")
```

```
GLMM6 <- manyglm(preymvabund ~ Season +(1|IDModel), data = GLMMGCol, family =  
"negative.binomial")
```

```
drop1(GLMM6, test = "Chisq")
```

```
anova.manyglm(GLMM6)
```

Lake maps and equipment location

```
require(sf)
```

```
library(dplyr)
```

```
library(ggplot2)
```

```
library(ggrepel)
```

```
library(tidyr)
```

```
library(tidyverse)
```

```
library(patchwork)
```

```
library(gridExtra)
```

```
library(cowplot)
```

```
LATDMtoDDcoordinate <- function(coordinate) {
```

```
  split_coordinate <- strsplit(coordinate, " ")[[1]]
```

```
  # Extract degrees, minutes, and seconds from the latitude
```

```
  latitude_parts <- strsplit(split_coordinate[1], "°")[[1]]
```

```
  latitude_deg <- as.numeric(latitude_parts[1])
```

```
  latitude_min_parts <- strsplit(latitude_parts[2], " ")
```

```
  latitude_min <- as.numeric(latitude_min_parts[1])
```

```
  # Calculate the latitude in decimal degrees
```

```
  latitude <- latitude_deg + latitude_min / 60
```

```
  hemisphere <- split_coordinate[2]
```

```
  latitude <- as.numeric(latitude)
```

```
  return(latitude)
```

```
}
```

```
LOTDMtoDDcoordinate <- function(coordinate) {
```

```
  split_coordinate <- strsplit(coordinate, " ")[[1]]
```

```
  # Extract degrees, minutes, and seconds from the latitude
```

```
  latitude_parts <- strsplit(split_coordinate[1], "°")[[1]]
```

```
  latitude_deg <- as.numeric(latitude_parts[1])
```

```
  latitude_min_parts <- strsplit(latitude_parts[2], " ")
```

```
  latitude_min <- as.numeric(latitude_min_parts[1])
```

```
  # Calculate the latitude in decimal degrees
```

```
  longitude <- latitude_deg + latitude_min / 60
```

```
  hemisphere <- split_coordinate[2]
```

```
  # Determine the hemisphere and adjust the sign of the longitude
```

```
  if (tolower(split_coordinate[2]) == "w") {
```

```

longitude <- -longitude
}

longitude_numeric <- as.numeric(longitude)
return(longitude_numeric)
}

#####
#datasets
#####

gaspe.df <- read.csv("~/Documents/BiologiMaster/Masteroppgave
/Excel/CSV_files/equipment_locations.csv", sep=";")

gaspe.df$latDD <- lapply(gaspe.df$lat_A, LATDMtoDDcoordinate)

gaspe.df$lonDD <- lapply(gaspe.df$long_A, LOTDMtoDDcoordinate)

#####
#Cascapedia summer
#####

setwd("~/Documents/BiologiMaster/Masteroppgave /KML.files")
cascSpace <- st_read("cascapedia.kml") %>%
  st_transform(32620)

cascSpace <- cascSpace %>%
  slice(1) %>%
  st_difference(cascSpace %>% slice(2)) %>%
  st_difference(cascSpace %>% slice(3)) %>%
  st_difference(cascSpace %>% slice(4))

urm <- 32620

```

```
cas_selected <- dplyr::select(cascSpace, geometry) %>% st_zm()
```

```
cas.df <- subset(gaspe.df, lake=="Cascapedia")
```

```
casS23.df <- subset(cas.df, Season=="S23")
```

```
casNet.df <- subset(casS23.df, type=="NET")
```

```
casNet_tibble.df <- as_tibble(casNet.df)
```

```
casNet_tibble.df <- dplyr::filter(casNet_tibble.df, !is.na(lonDD) & !is.na(latDD))
```

```
casNet_space.df <- st_as_sf(casNet_tibble.df, coords = c("lonDD", "latDD"))
```

```
casNet_space.df <- st_set_crs(casNet_space.df, 4326)
```

```
connections_df <- data.frame(
```

```
  from = c(1, 2, 3), # Index of the starting points in sf_object
```

```
  to = c(4, 5, 6)   # Index of the ending points in sf_object
```

```
)
```

```
line <- st_sfc(st_linestring(st_coordinates(casNet_space.df)),
```

```
  crs = st_crs(casNet_space.df))
```

```
allCoords <- as.matrix(st_coordinates(casNet_space.df))
```

```
lines <- lapply(1:nrow(connections_df),
```

```
  function(r){
```

```
    rbind(allCoords[connections_df[r,1], ],
```

```
          allCoords[connections_df[r,2], ])
```

```
  }) %>%
```

```
  st_multilinestring(.) %>%
```

```
  st_sfc(., crs = st_crs(casNet_space.df))
```

```
# Select the 'geometry' column from 'th' and set Z and M values
```

```
cas_selected <- dplyr::select(cascSpace, geometry) %>% st_zm()
```



```

cascS23_plot <- ggplot() +
  geom_sf(data = casc_selected, color="#03071e", fill="#a1b5d8") +
  geom_sf(data = cascNet_space.df, aes(color = "cascNet_space", shape = "cascNet_space"),
show.legend = TRUE) +
  geom_sf(data = lines, color = "#03071e", linetype="longdash") +
  theme(panel.grid = element_blank(),
        axis.text.x= element_blank(),
        axis.text.y= element_blank(),
        panel.background = element_rect(fill = "transparent", color = NA),
        axis.ticks.x = element_blank(),
        axis.ticks.y = element_blank(),
        legend.key = element_rect(fill = "transparent"),
        plot.margin = unit(c(0.5,0.5,0.5,0.5), "cm"),
        legend.text = element_text(size=8),
        #legend.position = c(0.05, .95)
) +
#legend.justification = c("right", "bottom"))
scale_color_manual(name = "Sample equipment",
  values = c("#4f772d"),
  labels = c("Nets")) +
scale_shape_manual(name = "Sample equipment",
  values = c(16),
  labels = c("Nets"))

# Add scale and North arrow
cascS23_plot <- cascS23_plot+
  ggspatial::annotation_scale(
    location = "br",
    bar_cols = c("#c2d8b9", "#738290"),
  ) +
  ggspatial::annotation_north_arrow(
    location = "tl", which_north = "true",
    pad_x = unit(0.4, "in"), pad_y = unit(0.4, "in"),
    style = ggspatial::north_arrow_fancy_orientteering(

```

```

    fill = c("#c2d8b9", "#738290"),
    line_col = "grey20",
  )
)

print(cascS23_plot)

#####
#Haymard summer
#####
HaySpace <- st_read("Haymard.KML") %>%
  st_transform(32620)

HaySpace <- HaySpace %>%
  slice(1) %>%
  st_difference(HaySpace %>% slice(2)) %>%
  st_difference(HaySpace %>% slice(3)) %>%
  st_difference(HaySpace %>% slice(4))
urm <- 32620

Hay_selected <- dplyr::select(HaySpace, geometry) %>% st_zm()
Hay.df <- subset(gaspe.df, lake=="Haymard")
HayS23.df <- subset(Hay.df, Season=="S23")
HayNetS23.df <- subset(HayS23.df, type=="NET")

HayNetS23_tibble.df <- as_tibble(HayNetS23.df)
HayNetS23_tibble.df <- dplyr::filter(HayNetS23_tibble.df, !is.na(lonDD) & !is.na(latDD))
HayNetS23_space.df <- st_as_sf(HayNetS23_tibble.df, coords = c("lonDD", "latDD"))
HayNetS23_space.df <- st_set_crs(HayNetS23_space.df, 4326)

# Select the 'geometry' column from 'th' and set Z and M values
Hay_selected <- dplyr::select(HaySpace, geometry) %>% st_zm()

```

```

connections_df <- data.frame(
  from = c(1, 2, 3), # Index of the starting points in sf_object
  to = c(4, 5, 6)   # Index of the ending points in sf_object
)

line <- st_sfc(st_linestring(st_coordinates(HayNetS23_space.df)),
  crs = st_crs(HayNetS23_space.df))

allCoords <- as.matrix(st_coordinates(HayNetS23_space.df))
lines <- lapply(1:nrow(connections_df),
  function(r){
    rbind(allCoords[connections_df[r,1], ],
      allCoords[connections_df[r,2], ])
  }) %>%
  st_multilinestring(.) %>%
  st_sfc(., crs = st_crs(HayNetS23_space.df))

#####
#Haymard plot
#####
HayS23_plot <- ggplot() +
  geom_sf(data = Hay_selected , color="#03071e", fill="#a1b5d8") +
  geom_sf(data = HayNetS23_space.df, aes(color = "HayNet_space", shape =
"HayNet_space"), show.legend = TRUE) +
  geom_sf(data = lines, color = "#03071e", linetype="solid")+
  theme(panel.grid = element_blank(),
    axis.text.x= element_blank(),
    axis.text.y= element_blank(),
    panel.background = element_rect(fill = "transparent", color = NA),
    axis.ticks.x = element_blank(),
    axis.ticks.y = element_blank(),
    legend.key = element_rect(fill = "transparent"),
    plot.margin = unit(c(0.5,0.5,0.5,0.5), "cm")) +
  scale_color_manual(name = "Sample equipment",

```

```

        values = c("#4f772d"),
        labels = c("Nets")) +
scale_fill_manual(name = "Sample equipment",
        values = c("#4f772d"),
        labels = c("Nets")) +
scale_shape_manual(name = "Sample equipment",
        values = c(16),
        labels = c("Nets","Traps"))

HayS23_plot <- HayS23_plot+
  ggspatial::annotation_scale(
    location = "br",
    bar_cols = c("#c2d8b9", "#738290"),
  ) +
  ggspatial::annotation_north_arrow(
    location = "tr", which_north = "true",
    pad_x = unit(1.5, "in"), pad_y = unit(0, "in"),
    style = ggspatial::north_arrow_fancy_orienteering(
      fill = c("#c2d8b9", "#738290"),
      line_col = "grey20",
    )
  )

print(HayS23_plot)

#####
#Paul summer
#####

paulSpace <- st_read("Paul.KML") %>%
  st_transform(32620)

paulSpace <- paulSpace %>%

```

```

slice(1) %>%
st_difference(paulSpace %>% slice(2)) %>%
st_difference(paulSpace %>% slice(3)) %>%
st_difference(paulSpace %>% slice(4))

urm <- 32620

paul_selected <- dplyr::select(paulSpace, geometry) %>% st_zm()

Paul.df <- subset(gaspe.df, lake=="Paul")
PaulS23.df <- subset(Paul.df, Season=="S23")
PaulNet.df <- subset(PaulS23.df, type=="NET")

PaulNet_tibble.df <- as_tibble(PaulNet.df)
PaulNet_tibble.df <- dplyr::filter(PaulNet_tibble.df, !is.na(lonDD) & !is.na(latDD))
PaulNet_space.df <- st_as_sf(PaulNet_tibble.df, coords = c("lonDD", "latDD"))
PaulNet_space.df <- st_set_crs(PaulNet_space.df, 4326)

#####
#Lines
#####
connections_df <- data.frame(
  from = c(1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 31, 32, 33), # Index of the starting
points in sf_object
  to = c(16, 17, 18, 19, 20,21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 34, 35, 36) # Index of the
ending points in sf_object

)

line <- st_sfc(st_linestring(st_coordinates(PaulNet_space.df)),
  crs = st_crs(PaulNet_space.df))

allCoords <- as.matrix(st_coordinates(PaulNet_space.df))
lines <- lapply(1:nrow(connections_df),

```

```

function(r){
  rbind(allCoords[connections_df[r,1], ],
        allCoords[connections_df[r,2], ])
  }) %>%
st_multilinestring(.) %>%
st_sfc(., crs = st_crs(PaulNet_space.df))

#####
#Paulplot
#####
paul_selected <- dplyr::select(paulSpace, geometry) %>% st_zm()

paulS23_plot <- ggplot() +
  geom_sf(data = paul_selected, color="#03071e", fill="#a1b5d8") +
  geom_sf(data = PaulNet_space.df, aes(color = "PaulNet_space", shape = "PaulNet_space"),
show.legend = TRUE) +
  geom_sf(data = lines, color = "#03071e", linetype="solid")+
  theme(panel.grid = element_blank(),
        axis.text.x= element_blank(),
        axis.text.y= element_blank(),
        panel.background = element_rect(fill = "transparent", color = NA),
        axis.ticks.x = element_blank(),
        axis.ticks.y = element_blank(),
        legend.key = element_rect(fill = "transparent"),
        plot.margin = unit(c(0,0,0,0), "cm"))+
  scale_color_manual(name = "Sample equipment",
                    values = c("#4f772d"),
                    labels = c("Nets")) +
  scale_fill_manual(name = "Sample equipment",
                   values = c( "#4f772d" ),
                   labels = c("Nets")) +
  scale_shape_manual(name = "Sample equipment",
                    values = c(16),
                    labels = c("Nets"))

```

```

paulS23_plot <- paulS23_plot+
  ggspatial::annotation_scale(
    location = "tr",
    pad_x = unit(0.1, "in"), pad_y = unit(0.8, "in"),
    bar_cols = c("#c2d8b9", "#738290"),
  )+
  ggspatial::annotation_north_arrow(
    location = "tr", which_north = "true",
    pad_x = unit(0.4, "in"), pad_y = unit(0.1, "in"),
    style = ggspatial::north_arrow_fancy_orienteering(
      fill = c("#c2d8b9", "#738290"),
      line_col = "grey20",
    )
  )
)

print(paulS23_plot)

#####
#Thibault summer
#####

thibSpace <- st_read("thibault.KML") %>%
  st_transform(32620)

step1 <- thibSpace %>% dplyr::slice(1)
step2 <- step1 %>% st_difference(thibSpace %>% dplyr::slice(2))
step3 <- step2 %>% st_difference(thibSpace %>% dplyr::slice(3))
step4 <- step3 %>% st_difference(thibSpace %>% dplyr::slice(4))

valid_geometries <- st_is_valid(thibSpace$geometry)
invalid_geometries <- which(!valid_geometries)
print(paste("Invalid geometries at indices: ", paste(invalid_geometries, collapse = ", ")))

urm <- 32620

```

```

Thib_selected <- dplyr::select(thibSpace, geometry) %>% st_zm()
Thibault.df <- subset(gaspe.df, lake=="Thibault")
ThibaultS23.df <- subset(Thibault.df, Season=="S23")
ThibaultNet.df <- subset(ThibaultS23.df, type=="NET")

ThibaultNet_tibble.df <- as_tibble(ThibaultNet.df)
ThibaultNet_tibble.df <- dplyr::filter(ThibaultNet_tibble.df, !is.na(lonDD) & !is.na(latDD))
ThibaultNet_space.df <- st_as_sf(ThibaultNet_tibble.df, coords = c("lonDD", "latDD"))
ThibaultNet_space.df <- st_set_crs(ThibaultNet_space.df, 4326)

connections_df <- data.frame(
  from = c(1, 2, 3, 4, 5, 6, 7, 8, 9, 10), # Index of the starting points in sf_object
  to = c(11, 12, 13, 14, 15, 16, 17, 18, 19, 20) # Index of the ending points in sf_object
)

line <- st_sfc(st_linestring(st_coordinates(ThibaultNet_space.df)),
  crs = st_crs(ThibaultNet_space.df))

allCoords <- as.matrix(st_coordinates(ThibaultNet_space.df))
lines <- lapply(1:nrow(connections_df),
  function(r){
    rbind(allCoords[connections_df[r,1], ],
      allCoords[connections_df[r,2], ])
  }) %>%
  st_multilinestring(.) %>%
  st_sfc(., crs = st_crs(ThibaultNet_space.df))
plot(lines)

thibS23_plot <- ggplot() +
  geom_sf(data = Thib_selected, color="#03071e", fill="#a1b5d8") +
  geom_sf(data = ThibaultNet_space.df, aes(color = "ThibaultNet_space", shape =
"ThibaultNet_space"), show.legend = TRUE) +
  geom_sf(data = lines, color = "#03071e", linetype="solid") +

```



```

theme(panel.grid = element_blank(),
      axis.text.x= element_blank(),
      axis.text.y= element_blank(),
      panel.background = element_rect(fill = "transparent", color = NA, size = 2),
      axis.ticks.x = element_blank(),
      axis.ticks.y = element_blank(),
      legend.key = element_rect(fill = "transparent"),
      plot.margin = unit(c(0.5,0.5,0.5,0.5), "cm"))+
scale_color_manual(name = "Sample equipment",
                   values = c("#4f772d"),
                   labels = c("Nets")) +
scale_fill_manual(name = "#4f772d",
                  values = c("EC600A" ),
                  labels = c("Nets")) +
scale_shape_manual(name = "Sample equipment",
                   values = c(16),
                   labels = c("Nets"))# Add scale and North arrow

```

```

thibS23_plot <- thibS23_plot+
ggspatial::annotation_scale(
  location = "br",
  bar_cols = c("#c2d8b9", "#738290"),
) +
ggspatial::annotation_north_arrow(
  location = "tr", which_north = "true",
  pad_x = unit(0, "in"), pad_y = unit(0.1, "in"),
  style = ggspatial::north_arrow_fancy_orientteering(
    fill = c("#c2d8b9", "#738290"),
    line_col = "grey20",
  )
)

```

```

print(thibS23_plot)

```

```

#####
#Cascapedia winter
#####
caseSpace <- st_read("cascapedia.KML") %>%
  st_transform(32620)

caseSpace <- caseSpace %>%
  slice(1) %>%
  st_difference(caseSpace %>% slice(2)) %>%
  st_difference(caseSpace %>% slice(3)) %>%
  st_difference(caseSpace %>% slice(4))

urm <- 32620

case_selected <- dplyr::select(caseSpace, geometry) %>% st_zm()

#####
#casplot
#####
case.df <- subset(gaspe.df, lake=="Cascapedia")
caseW23.df <- subset(case.df, Season=="W23")
caseTU.df <- subset(caseW23.df, type=="TU")
caseTrap.df <- subset(caseW23.df, type=="trap")

#####
#convert TU
#####
caseTU_tibble.df <- as_tibble(caseTU.df)
caseTU_tibble.df <- dplyr::filter(caseTU_tibble.df, !is.na(lonDD) & !is.na(latDD))
caseTU_space.df <- st_as_sf(caseTU_tibble.df, coords = c("lonDD", "latDD"))
caseTU_space.df <- st_set_crs(caseTU_space.df, 4326)

#####

```

```

#convert traps
#####
cascTrap_tibble.df <- as_tibble(cascTrap.df)
cascTrap_tibble.df <- dplyr::filter(cascTrap_tibble.df, !is.na(lonDD) & !is.na(latDD))
cascTrap_space.df <- st_as_sf(cascTrap_tibble.df, coords = c("lonDD", "latDD"))
cascTrap_space.df <- st_set_crs(cascTrap_space.df, 4326)

# Select the 'geometry' column from 'th' and set Z and M values
casc_selected <- dplyr::select(cascSpace, geometry) %>% st_zm()

cascW23_plot <- ggplot() +
  geom_sf(data = casc_selected, color="#03071e", fill="#a1b5d8") +
  geom_sf(data = cascTrap_space.df, aes(color = "cascTrap_space", shape =
"cascTrap_space"), show.legend = TRUE) +
  geom_sf(data = cascTU_space.df, aes(color = "cascTU_space", shape = "cascTU_space"),
show.legend = TRUE) +
  #geom_path(data = ThibaultNet_space.df, aes(x = your_x_column, y = your_y_column,
group = group_column), color = "blue") + # Replace your_x_column, your_y_column, and
group_column with appropriate column names
  theme(panel.grid = element_blank(),
        axis.text.x= element_blank(),
        axis.text.y= element_blank(),
        panel.background = element_rect(fill = "transparent", color = NA),
        axis.ticks.x = element_blank(),
        axis.ticks.y = element_blank(),
        legend.key = element_rect(fill = "transparent"),
        plot.margin = unit(c(0.5,0.5,0.5,0.5), "cm")) +
  #legend.text = element_text(size=8),
  #legend.position = c(0.05, .95),
  #legend.justification = c("right", "bottom"))
scale_color_manual(name = "Sample equipment",
  values = c("#132a13", "#ffe169"),
  labels = c("Traps", "Tip-Ups")) +
scale_fill_manual(name = "Sample equipment",

```

```

        values = c("#132a13", "#ffe169" ),
        labels = c("Traps", "Tip-Ups")) +
scale_shape_manual(name = "Sample equipment",
        values = c(15, 88),
        labels = c("Traps", "Tip-Ups"))

# Add scale and North arrow
caseW23_plot <- caseW23_plot+
  ggspatial::annotation_scale(
    location = "br",
    bar_cols = c("#c2d8b9", "#738290"),
    text_family = "ArcherPro Book"
  ) +
  ggspatial::annotation_north_arrow(
    location = "tl", which_north = "true",
    pad_x = unit(0.4, "in"), pad_y = unit(0.4, "in"),
    style = ggspatial::north_arrow_fancy_orienteeing(
      fill = c("#c2d8b9", "#738290"),
      line_col = "grey20",
      text_family = "ArcherPro Book"
    )
  )

print(caseW23_plot)

#####
#Paul winter
#####
paulSpace <- st_read("Paul.KML") %>%
  st_transform(32620)

paulSpace <- paulSpace %>%
  slice(1) %>%

```

```
st_difference(paulSpace %>% slice(2)) %>%  
st_difference(paulSpace %>% slice(3)) %>%  
st_difference(paulSpace %>% slice(4))
```

```
urm <- 32620
```

```
paul_selected <- dplyr::select(paulSpace, geometry) %>% st_zm()
```

```
Paul.df <- subset(gaspe.df, lake=="Paul")  
PaulW23.df <- subset(Paul.df, Season=="W23")  
PaulTU.df <- subset(PaulW23.df, type=="TU")  
PaulNet.df <- subset(PaulW23.df, type=="NET")
```

```
#####
```

```
#convert TU
```

```
#####
```

```
PaulTU_tibble.df <- as_tibble(PaulTU.df)  
PaulTU_tibble.df <- dplyr::filter(PaulTU_tibble.df, !is.na(lonDD) & !is.na(latDD))  
PaulTU_space.df <- st_as_sf(PaulTU_tibble.df, coords = c("lonDD", "latDD"))  
PaulTU_space.df <- st_set_crs(PaulTU_space.df, 4326)
```

```
#####
```

```
#convert nets
```

```
#####
```

```
PaulNet_tibble.df <- as_tibble(PaulNet.df)  
PaulNet_tibble.df <- dplyr::filter(PaulNet_tibble.df, !is.na(lonDD) & !is.na(latDD))  
PaulNet_space.df <- st_as_sf(PaulNet_tibble.df, coords = c("lonDD", "latDD"))  
PaulNet_space.df <- st_set_crs(PaulNet_space.df, 4326)
```

```
#####
```

```
#lines
```

```

#####
connections_df <- data.frame(
  from = c(1, 2, 3, 4, 5), # Index of the starting points in sf_object
  to = c(6, 7, 8, 9, 10)  # Index of the ending points in sf_object
)

line <- st_sfc(st_linestring(st_coordinates(cascNet_space.df)),
  crs = st_crs(PaulNet_space.df))

allCoords <- as.matrix(st_coordinates(PaulNet_space.df))
lines <- lapply(1:nrow(connections_df),
  function(r){
    rbind(allCoords[connections_df[r,1], ],
      allCoords[connections_df[r,2], ])
  }) %>%
  st_multilinestring(.) %>%
  st_sfc(., crs = st_crs(PaulNet_space.df))

#Plot
paul_selected <- dplyr::select(paulSpace, geometry) %>% st_zm()

paulW23_plot <- ggplot() +
  geom_sf(data = paul_selected, color="#03071e", fill="#a1b5d8") +
  geom_sf(data = PaulNet_space.df, aes(color = "PaulNet_space", shape = "PaulNet_space"),
  show.legend = TRUE) +
  geom_sf(data = PaulTU_space.df, aes(color = "PaulTU_space", shape = "PaulTU_space"),
  show.legend = TRUE) +
  geom_sf(data = lines, color = "#03071e", linetype="solid")+
  theme(panel.grid = element_blank(),
    axis.text.x= element_blank(),
    axis.text.y= element_blank(),
    panel.background = element_rect(fill = "transparent", color = NA),
    axis.ticks.x = element_blank(),

```

```

axis.ticks.y = element_blank(),
legend.key = element_rect(fill = "transparent"),
plot.margin = unit(c(0,0,0,0), "cm") +
scale_color_manual(name = "Sample equipment",
  values = c("#4f772d", "#ffe169"),
  labels = c("Nets", "Tip-Ups")) +
scale_fill_manual(name = "Sample equipment",
  values = c("#4f772d", "#ffe169"),
  labels = c("Nets", "Tip-Ups")) +
scale_shape_manual(name = "Sample equipment",
  values = c(16, 88),
  labels = c("Nets", "Tip-Ups"))

# Add scale and North arrow
paulW23_plot <- paulW23_plot+
ggspatial::annotation_scale(
  location = "br", pad_x = unit(0.1, "in"), pad_y = unit(0, "in"),
  bar_cols = c("#c2d8b9", "#738290"),
  text_family = "ArcherPro Book"
) +
ggspatial::annotation_north_arrow(
  location = "tr", which_north = "true",
  pad_x = unit(0, "in"), pad_y = unit(0.2, "in"),
  style = ggspatial::north_arrow_fancy_orientteering(
    fill = c("#c2d8b9", "#738290"),
    line_col = "grey20",
    text_family = "ArcherPro Book"
  )
)

print(paulW23_plot)

#####
#Thibault winter

```

```
#####
```

```
thibSpace <- st_read("thibault.kml") %>%  
  st_transform(32620)
```

```
step1 <- thibSpace %>% dplyr::slice(1)  
step2 <- step1 %>% st_difference(thibSpace %>% dplyr::slice(2))  
step3 <- step2 %>% st_difference(thibSpace %>% dplyr::slice(3))  
step4 <- step3 %>% st_difference(thibSpace %>% dplyr::slice(4))
```

```
valid_geometries <- st_is_valid(thibSpace$geometry)  
invalid_geometries <- which(!valid_geometries)  
print(paste("Invalid geometries at indices: ", paste(invalid_geometries, collapse = ", ")))  
urm <- 32620
```

```
Thib_selected <- dplyr::select(thibSpace, geometry) %>% st_zm()
```

```
Thibault.df <- subset(gaspe.df, lake=="Thibault")  
ThibaultW23.df <- subset(Thibault.df, Season=="W23")  
ThibaultTU.df <- subset(ThibaultW23.df, type=="TU")  
ThibaultTrap.df <- subset(ThibaultW23.df, type=="trap")
```

```
ThibaultTU_tibble.df <- as_tibble(ThibaultTU.df)  
ThibaultTU_tibble.df <- dplyr::filter(ThibaultTU_tibble.df, !is.na(lonDD) & !is.na(latDD))  
ThibaultTU_space.df <- st_as_sf(ThibaultTU_tibble.df, coords = c("lonDD", "latDD"))  
ThibaultTU_space.df <- st_set_crs(ThibaultTU_space.df, 4326)
```

```
ThibaultTrap_tibble.df <- as_tibble(ThibaultTrap.df)  
ThibaultTrap_tibble.df <- dplyr::filter(ThibaultTrap_tibble.df, !is.na(lonDD) & !is.na(latDD))  
ThibaultTrap_space.df <- st_as_sf(ThibaultTrap_tibble.df, coords = c("lonDD", "latDD"))  
ThibaultTrap_space.df <- st_set_crs(ThibaultTrap_space.df, 4326)
```

```
thibW23_plot <- ggplot() +  
  geom_sf(data = Thib_selected, color="#03071e", fill="#a1b5d8") +
```



```

geom_sf(data = ThibaultTU_space.df, aes(color = "ThibaultTU_space", shape =
"ThibaultTU_space"), show.legend = TRUE) +
geom_sf(data = ThibaultTrap_space.df, aes(color = "ThibaultTrap_space", shape =
"ThibaultTrap_space"), show.legend = TRUE) +
theme(panel.grid = element_blank(),
      axis.text.x= element_blank(),
      axis.text.y= element_blank(),
      panel.background = element_rect(fill = "transparent", color = NA, size = 2),
      axis.ticks.x = element_blank(),
      axis.ticks.y = element_blank(),
      #panel.border = element_rect(color = "black",
      #fill = NA,
      #linewidth = 2),
      legend.key = element_rect(fill = "transparent"),
      plot.margin = unit(c(0.5,0.5,0.5,0.5), "cm"))+
#legend.text = element_text(size=8),
#legend.position = c(0.05, .95),
#legend.justification = c("right", "bottom"))
scale_color_manual(name = "Sample equipment",
                   values = c("#132a13", "#ffe169"),
                   labels = c("Traps", "Tip-Ups")) +
scale_fill_manual(name = "Sample equipment",
                  values = c("#132a13", "#ffe169" ),
                  labels = c("Traps", "Tip-Ups")) +
scale_shape_manual(name = "Sample equipment",
                   values = c(15, 88),
                   labels = c("Traps", "Tip-Ups"))# Add scale and North arrow

thibW23_plot <- thibW23_plot+
ggspatial::annotation_scale(
  location = "br",
  bar_cols = c("#c2d8b9", "#738290"),
  text_family = "ArcherPro Book"
) +

```

```

ggspatial::annotation_north_arrow(
  location = "tr", which_north = "true",
  pad_x = unit(0, "in"), pad_y = unit(0.1, "in"),
  style = ggspatial::north_arrow_fancy_orienteeing(
    fill = c("#c2d8b9", "#738290"),
    line_col = "grey20",
    text_family = "ArcherPro Book"
  )
)
)

```

```
print(thibW23_plot)
```

RINSP analysis

```

library(RInSp)
library(vegan)
library(dplyr)

```

```

setwd("~/Documents/BiologiMaster/Masteroppgave /Excel/CSV_files")
safo243 <- read.csv2("~/Documents/BiologiMaster/Masteroppgave
/Excel/CSV_files/safo243.csv")

```

```

safo <- safo243 %>%
  mutate_at(vars(5:29), as.numeric)

```

```

winter <- subset(safo, Season=="Winter")
summer <- subset(safo, Season=="Summer")

```

```

#####
#winter
#####

```

```

#Paul
safopauRISWi = import.RInSp(winter, col.header=TRUE, row.names=1, info.cols=c(1:4),
data.type = "integer", subset.rows = c("Lake", "Paul"))
resultsPauWi <- WTcMC(safopauRISWi, replicates = 999)
resultsPauWWi= WTcMC(safopauRISWi, weight="N_items", replicates=999)
TroutEPauWi = Eindex(safopauRISWi, index = "saramaki", jackknife = TRUE)
sumMC.RInSp(resultsPauWWi)
PSiPauWi <- PSicalc(safopauRISWi, exclude = FALSE, replicates = 999)

```

```

#Cascapedia
safocascRISWi = import.RInSp(winter, col.header=TRUE, row.names=1, info.cols=c(1:4),
data.type = "integer", subset.rows = c("Lake", "Cascapedia"))
resultsCascWi <- WTcMC(safocascRISWi, replicates = 999)

```

```
resultsCascWWi= WTcMC(safocascRISWi, weight="N_items", replicates=999)
TroutECascWi = Eindex(safocascRISWi, index = "saramaki", jackknife = TRUE)
sumMC.RInSp(resultsCascWWi)
PSiCascWi <- PSicalc(safocascRISWi, exclude = FALSE, replicates = 999)
```

```
#Thibault
```

```
safothibRISWi = import.RInSp(winter, col.header=TRUE, row.names=1, info.cols=c(1:4),
data.type = "integer", subset.rows = c("Lake", "Thibault"))
resultsThibWi <- WTcMC(safothibRISWi, replicates = 999)
resultsThibWWi = WTcMC(safothibRISWi, weight="N_items", replicates=999)
TroutEWi = Eindex(safothibRISWi, index = "saramaki", jackknife = TRUE)
sumMC.RInSp(resultsThibWWi)
PSiThibWi <- PSicalc(safothibRISWi, exclude = FALSE, replicates = 999)
```

```
#####
```

```
#summer
```

```
#####
```

```
#Paul
```

```
safopauRISSu = import.RInSp(summer, col.header=TRUE, row.names=1, info.cols=c(1:4),
data.type = "integer", subset.rows = c("Lake", "Paul"))
resultsPauSu <- WTcMC(safopauRISSu, replicates = 999)
resultsPauWSu= WTcMC(safopauRISSu, weight="N_items", replicates=999)
TroutEPauSu = Eindex(safopauRISSu, index = "saramaki", jackknife = TRUE)
sumMC.RInSp(resultsPauSu)
PSipauSu <- PSicalc(safopauRISSu, exclude = FALSE, replicates = 999)
```

```
#Cascapedia
```

```
safocascRISSu = import.RInSp(summer, col.header=TRUE, row.names=1, info.cols=c(1:4),
data.type = "integer", subset.rows = c("Lake", "Cascapedia"))
resultsCascSu <- WTcMC(safocascRISSu, replicates = 999)
resultsCascWSu= WTcMC(safocascRISSu, weight="N_items", replicates=999)
TroutECascSu = Eindex(safocascRISSu, index = "saramaki", jackknife = TRUE)
sumMC.RInSp(resultsCascWSu)
PSicascSu <- PSicalc(safocascRISSu, exclude = FALSE, replicates = 999)
```

```
#Thibault
```

```
safothibRISSu = import.RInSp(summer, col.header=TRUE, row.names=1, info.cols=c(1:4),
data.type = "integer", subset.rows = c("Lake", "Thibault"))
resultsThibWi <- WTcMC(safothibRISSu, replicates = 999)
resultsThibWSu = WTcMC(safothibRISSu, weight="N_items", replicates=999)
TroutESu = Eindex(safothibRISSu, index = "saramaki", jackknife = TRUE)
sumMC.RInSp(resultsThibWSu)
PSiThibSu <- PSicalc(safothibRISSu, exclude = FALSE, replicates = 999)
```

```
#Haymard
```

```
safohayRISSu = import.RInSp(summer, col.header=TRUE, row.names=1, info.cols=c(1:4),
data.type = "integer", subset.rows = c("Lake", "Haymard"))
resultsHay <- WTcMC(safohayRISSu, replicates = 999)
resultsHayWSu= WTcMC(safohayRISSu, weight="N_items", replicates=999)
safohayESu = Eindex(safohayRISSu, index = "saramaki", jackknife = TRUE)
```

```

sumMC.RInSp(resultsHayWSu)
PSiHaySu <- PSicalc(safoHayRISSu, exclude = FALSE, replicates = 999)

#####
#Shannon diversity index
#####

#####
#Summer
#####
#Haymard
summerHay <- subset(summer, Lake=="Haymard")
summerShannHay <- summerHay %>% select(c(-Date,-Season,-Lake))
shannonHaySum <- (mean(diversity(summerShannHay, index = "shannon")))
shannonHaySum

#Cascapedia
summerCasc <- subset(summer, Lake=="Cascapedia")
summerShannCasc <- summerCasc %>% select(c(-Date,-Season,-Lake))
shannonCascSum <- (mean(diversity(summerShannCasc, index = "shannon")))
shannonCascSum

#Paul
summerPaul <- subset(summer, Lake=="Paul")
summerShannPaul <- summerPaul %>% select(c(-Date,-Season,-Lake))
shannonPaulSum <- (mean(diversity(summerShannPaul, index = "shannon")))
shannonPaulSum

#Thibault
summerThib <- subset(summer, Lake=="Thibault")
summerShannThib <- summerThib %>% select(c(-Date,-Season,-Lake))
shannonTHibSum <- (mean(diversity(summerShannThib, index = "shannon")))
shannonTHibSum

#####
#Winter
#####
#Cascapedia
winterCasc <- subset(winter, Lake=="Cascapedia")
winterShannCasc <- winterCasc %>% select(c(-Date,-Season,-Lake))
shannonCascWin <- (mean(diversity(winterShannCasc, index = "shannon")))
shannonCascWin

#Paul
winterPaul <- subset(winter, Lake=="Paul")
winterShannPaul <- winterPaul %>% select(c(-Date,-Season,-Lake))
shannonPaulWin <- (mean(diversity(winterShannPaul, index = "shannon")))
shannonPaulWin

#Thibault

```

```

winterThib <- subset(winter, Lake=="Thibault")
winterShannThib <- winterThib %>% select(c(-Date,-Season,-Lake))
shannonThibWin <- (mean(diversity(winterShannThib, index = "shannon")))
shannonThibWin

#####
#Simpson diversity index
#####

#####
#summer
#####
#Haymard
summerHay <- subset(summer, Lake=="Haymard")
summerSimpHay <- summerHay %>% select(c(-Date,-Season,-Lake))
simpsonHaySum <- (mean(diversity(summerSimpHay, index = "simpson")))
simpsonHaySum

#Cascapedia
summerCasc <- subset(summer, Lake=="Cascapedia")
summerSimpCasc <- summerCasc %>% select(c(-Date,-Season,-Lake))
simpsonCascSum <- (mean(diversity(summerSimpCasc, index = "simpson")))
simpsonCascSum

#Paul
summerPaul <- subset(summer, Lake=="Paul")
summerSimpPaul <- summerPaul %>% select(c(-Date,-Season,-Lake))
simpsonPaulSum <- (mean(diversity(summerSimpPaul, index = "simpson")))
simpsonPaulSum

#Thibault
summerThib <- subset(summer, Lake=="Thibault")
summerSimpThib <- summerThib %>% select(c(-Date,-Season,-Lake))
simpsonThibSum <- (mean(diversity(summerSimpThib, index = "simpson")))
simpsonThibSum

#####
#Winter
#####
#Cascapedia
winterCasc <- subset(winter, Lake=="Cascapedia")
winterSimpCasc <- winterCasc %>% select(c(-Date,-Season,-Lake))
simpsonCascWin <- (mean(diversity(winterSimpCasc, index = "simpson")))
simpsonCascWin

#Paul
winterPaul <- subset(winter, Lake=="Paul")
winterSimpPaul <- winterPaul %>% select(c(-Date,-Season,-Lake))
simpsonPaulWin <- (mean(diversity(winterSimpPaul, index = "simpson")))

```

```
simpsonPaulWin
```

```
#Thibault
```

```
winterThib <- subset(winter, Lake=="Thibault")  
winterSimpThib <- winterThib %>% select(c(-Date,-Season,-Lake))  
simpsonThibWin <- (mean(diversity(winterSimpThib, index = "simpson")))  
simpsonThibWin
```

RINSP plots

```
library(ggplot2)  
library(readr)
```

```
Index <- read.csv2("~/Documents/BiologiMaster/Masteroppgave  
/Excel/CSV_files/Index_values.csv")  
#mutate_at(vars(3:11), as.numeric)
```

```
Overlap <- ggplot(Index, aes(x = Lake, y = Specialization, fill = Season)) +  
  geom_bar(stat = "identity", position = position_dodge()) +  
  theme_bw(base_size = 15) +  
  scale_fill_manual(values = c("#cfe0c3", "#005478")) +  
  scale_y_continuous(limits = c(0,1)) +  
  theme(plot.margin = unit(c(0, 0, 0, 0), "cm")) +  
  theme (  
    panel.grid.major = element_blank(),  
    panel.grid.minor = element_blank(),  
    panel.background = element_rect(fill = "transparent"),  
    plot.background = element_rect(fill = "transparent", color = NA),  
    axis.text = element_text(size = 12),  
    legend.text = element_text(color = "black", size = 12),  
    legend.title = element_text(size = 10),  
    legend.direction = "horizontal",  
    legend.position = "bottom"  
  ) +  
  labs (x = "Lake",  
        y = "Degree of overlap",  
        fill = "")
```

Overlap

```
E_Index <- ggplot(Index, aes(x = Lake, y = E_value, fill = Season)) +  
  geom_bar(stat = "identity", position = position_dodge()) +  
  theme_bw(base_size = 15) +  
  scale_fill_manual(values = c("#cfe0c3", "#005478")) +  
  scale_y_continuous(limits = c(0,1)) +  
  theme(plot.margin = unit(c(0, 0, 0, 0), "cm")) +  
  theme (  
    panel.grid.major = element_blank(),  
    panel.grid.minor = element_blank(),  
    panel.background = element_rect(fill = "transparent"),
```

```

plot.background = element_rect(fill = "transparent", color = NA),
axis.text = element_text(size = 12),
legend.text = element_text(color = "black", size = 12),
legend.title = element_text(size = 10),
legend.direction = "horizontal",
legend.position = "bottom"
) +
labs (x = "Lake",
      y = "Degree of inter-individual niche variation",
      fill = "")
E_Index

```

```

PSi <- ggplot(Index, aes(x = Lake, y = Schoener, fill = Season)) +
  geom_bar(stat = "identity", position = position_dodge()) +
  theme_bw(base_size = 15) +
  scale_fill_manual(values = c("#cfe0c3", "#005478")) +
  scale_y_continuous(limits = c(0,1)) +
  theme(plot.margin = unit(c(0, 0, 0, 0), "cm")) +
  theme (
    panel.grid.major = element_blank(),
    panel.grid.minor = element_blank(),
    panel.background = element_rect(fill = "transparent"),
    plot.background = element_rect(fill = "transparent", color = NA),
    axis.text = element_text(size = 12),
    legend.text = element_text(color = "black", size = 12),
    legend.title = element_text(size = 10),
    legend.direction = "horizontal",
    legend.position = "bottom"
  ) +
  labs (x = "Lake",
        y = "Degree of individual specialization (PSi)",
        fill = "")
PSi

```

```

Cws <- ggplot(Index, aes(x = Lake, y = Cws, fill = Season)) +
  geom_bar(stat = "identity", position = position_dodge()) +
  theme_bw(base_size = 15) +
  scale_fill_manual(values = c("#cfe0c3", "#005478")) +
  scale_y_continuous(limits = c(-0.11,1)) +
  theme(plot.margin = unit(c(0, 0, 0, 0), "cm")) +
  theme (
    panel.grid.major = element_blank(),
    panel.grid.minor = element_blank(),
    panel.background = element_rect(fill = "transparent"),
    plot.background = element_rect(fill = "transparent", color = NA),
    axis.text = element_text(size = 12),
    legend.text = element_text(color = "black", size = 12),
    legend.title = element_text(size = 10),
    legend.direction = "horizontal",
    legend.position = "bottom"
  )

```

```
) +
labs (x = "Lake",
      y = " Degree of clustering in the network",
      fill = "")
```

Cws

```
Shannon <- ggplot(Index, aes(x = Lake, y = Shannon, fill = Season)) +
  geom_bar(stat = "identity", position = position_dodge()) +
  theme_bw(base_size = 15) +
  scale_fill_manual(values = c("#cfe0c3", "#005478")) +
  scale_y_continuous(limits = c(0,1)) +
  theme(plot.margin = unit(c(0, 0, 0, 0), "cm")) +
  theme (
    panel.grid.major = element_blank(),
    panel.grid.minor = element_blank(),
    panel.background = element_rect(fill = "transparent"),
    plot.background = element_rect(fill = "transparent", color = NA),
    axis.text = element_text(size = 12),
    legend.text = element_text(color = "black", size = 12),
    legend.title = element_text(size = 10),
    legend.direction = "horizontal",
    legend.position = "bottom"
  ) +
labs (x = "Lake",
      y = "Shannon index value",
      fill = "")
```

Shannon

```
Simpson <- ggplot(Index, aes(x = Lake, y = Simpson, fill = Season)) +
  geom_bar(stat = "identity", position = position_dodge()) +
  theme_bw(base_size = 15) +
  scale_fill_manual(values = c("#cfe0c3", "#005478")) +
  scale_y_continuous(limits = c(0,1)) +
  theme(plot.margin = unit(c(0, 0, 0, 0), "cm")) +
  theme (
    panel.grid.major = element_blank(),
    panel.grid.minor = element_blank(),
    panel.background = element_rect(fill = "transparent"),
    plot.background = element_rect(fill = "transparent", color = NA),
    axis.text = element_text(size = 12),
    legend.text = element_text(color = "black", size = 12),
    legend.title = element_text(size = 10),
    legend.direction = "horizontal",
    legend.position = "bottom"
  ) +
labs (x = "Lake",
      y = "Simpson index value",
      fill = "")
```

Simpson



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