UNIVERSITY OF GOTHENBURG

DEPARTMENT OF BIOLOGICAL AND ENVIRONMENTAL SCIENCES

TEMPERATURE PREFERENCE OF BROOK TROUT (*SALVELINUS FONTINALIS*) AND BROWN TROUT (*SALMO TRUTTA*)

An experimental study



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Abstract

Invasive alien species threaten native ecosystems by outcompeting native species or by negatively altering the function of the natural ecosystem. In Sweden, the invasive brook trout (*Salvelinus fontinalis*) displace native brown trout (*Salmo trutta*) from their food and habitat niches. Because salmonids are ectothermic, water temperature is a critical factor for their survival. Generally, brook trout is more frequently detected in upper stretches of watercourses with colder water. The aim of this study was to investigate the thermal preferences of both species and determine whether a difference exists under a controlled laboratory setting. A temperature preference annuli (TPA) was used, allowing the fish to select their position along a thermal gradient. A weighted temperature index was calculated to represent individual temperature preference. The result indicated a significantly lower temperature preference for brook trout compared to brown trout ($p = 0.027$). Furthermore, there was a statistically significant interaction between body condition and species on the weighted temperature index ($p = 0.037$), this indicates that how the body condition impacts temperature preference differs between species. Additionally, diurnal temperature preference was more variable for brook trout than for brown trout as water temperature increased. Overall, brook trout had a more varied temperature preference than brown trout, suggesting a higher phenotypic plasticity and individually based choices. This study provides fundamental data for future research. These findings contribute to a better understanding of brook trout and brown trout ecology in Sweden, informing conservation efforts, brook trout management and ecosystem responses to climate change.

Sammanfattning

Invasiva främmande arter hotar inhemska ekosystem genom att sprida sig och skapa konkurrens. I Sverige tränger den invasiva bäckrödingen (*Salvelinus fontinalis*) bort den inhemska öringen (*Salmo trutta*) från dess födo-och livsmiljönischer. Laxfiskar är ektoterma vilket innebär att vattentemperatur är en avgörande faktor för deras överlevnad. Bäckröding påträffas oftare i de övre delarna av vattendrag med kallare vatten. Syftet med denna studie var att undersöka båda arternas temperaturpreferenser och avgöra om det finns en skillnad under kontrollerade laboratorieförhållande. En anordning kallad ”*temperature preference annuli*” (TPA) användes, vilket gjorde det möjligt för fiskarna att välja sin position längs en temperaturgradient. Därefter beräknades ett viktat temperaturindex för att representera varje individs preferens. Resultaten indikerade på en signifikant lägre temperaturpreferens hos bäckröding jämfört med öring ($p = 0,027$). Det fanns en statistisk signifikant interaktion mellan kroppskondition och det viktade temperatur medelvärdet ($p = 0,037$), vilket tyder på att effekten av kroppskondition på temperaturpreferensen skiljer sig åt mellan arterna. Vidare uppvisade bäckrödingen en större spridning i temperaturpreferens under dagen än öringen gjorde. Sammantaget hade bäckrödingen en större variation i sin temperaturpreferens, vilket skulle kunna antyda att de har en högre fenotypisk plasticitet och individbaserade val. Denna studie utgör ett viktigt underlag för framtida forskning. Resultatet bidrar till en ökad förståelse för ekologin hos bäckröding och öring i Sverige, vilket är värdefullt för naturvård, förvaltning av bäckröding samt förståelsen av ekosystemens respons på klimatförändringar.

1. Introduction

In my thesis, I will study the temperature preference of brook trout (*Salvelinus fontinalis*) and brown trout (*Salmo trutta*) using an experimental arena with a temperature gradient. I want to contribute to a better understanding of brook trout's spatial distribution patterns, which can ultimately inform and improve management strategies for controlling and removing invasive brook trout from freshwater systems in Sweden.

1.1 Invasive alien species

Invasive alien species are organisms that are introduced by humans outside their natural ranges and habitats (IPBES, 2023). An alien species becomes invasive when it spreads fast and causes damage to nature by outcompeting other species (Naturvårdsverket, 2025). As they establish and spread, they can change the behavior of individuals and ecological functions (Simon & Townsend, 2003), ultimately degrading native ecosystems. By spreading and outcompeting native species for habitat and resources (Vitousek et al., 1997), they pose a major threat to biodiversity (Naturvårdsverket, 2025). Furthermore, coexistence with invasive species can lead to the breakdown of adaptive capacity, leading to reduced fitness in exposed native populations (Zavorka et al., 2017).

1.2 Study species and competition

An example of this dynamic is the invasive brook trout (*Salvelinus fontinalis*) (SLU Artdatabanken, 2026a) that partly have outcompeted native brown trout (*Salmo trutta*) in headwater streams across Northern Europe (Korsu et al., 2007). Brook trout originates from North America and was introduced by humans in the late 18th century and has since established populations throughout Sweden (MacCrimmon et al., 1969). This shift could be partly due to brook trout's behavior of occupying a distinct habitat niche (Korsu et al., 2012). Additionally, brook trout negatively impact brown trout by restricting their habitat choice, reducing their growth, and narrowing their food niche (Höjesjö & Austad, 2026). In addition, brook trout have high flexibility in habitat utilization, allowing them to rapidly colonize empty niches (Höjesjö & Austad, 2026). This colonization speed is likely impacted by brook trout's early reproductive time and that they spawn 2-3 weeks earlier than brown trout (Cucherousset et al., 2008). Furthermore, they also have a shorter period from fertilization to hatching (Grande & Anderson, 1990). This might be some of the factors resulting in a decrease in brown trout population in Sweden (SLU Artdatabanken, 2026b).

Furthermore, it has been detected that brown trout living in the same water as brook trout (sympatry) has different behaviors than in streams with only brown trout (allopatry). For example, sympatric brown trout has shown to be more diurnal and consumes more terrestrial prey than allopatric brown trout (Zavorka et al., 2017). In addition, Zavorka et al. (2019) demonstrated that sympatric brown trout has a divergence in head shape compared to allopatric brown trout because of niche convergence. Native species often fail to adapt their behavior and morphology to the extent required to coexist with biological invaders, such as invasive species, which can lead to long-term decreases in their population size (Lau & Terhorst, 2015). However, rather than being driven by direct aggressiveness between the species, Korsu et al. (2009) suggests that the success of brook trout may be attributed to habitat specialization. Brook trout occupies calm pools while brown trout utilize faster flowing waters, allowing them to establish successfully without any direct physical competition (Korsu et al., 2009).

1.3 Thermoregulation

For salmonids, water temperature is a fundamental factor affecting metabolic processes and therefore driving habitat selection (Jobling, 1981). Most fish species are ectothermic and rely on behavioral thermoregulation to occupy optimal thermal niches within their habitat (Magnuson et al., 1997). Being ectothermic means the body temperature is regulated by its environment, and changes in water temperature can directly affect physiological processes at the molecular, cellular, organ, and whole-organism levels (Ern et al., 2023). This means that they actively move to maintain their body temperature within their preference range when the surrounding temperature changes (Magnuson et al., 1997). In several species, seeking optimal temperature serves as an efficient strategy to increase growth rates (Jobling, 1981). Furthermore, thermal preferences and tolerance in salmonids can vary among individuals and life stage. In both brook trout and brown trout, factors such as body size, age, acclimation history, and environmental conditions influence how fish respond to water temperature (Elliot, 2013; Smith et al., 2020). Previous research on salmonids has shown that body condition can influence responses to thermal stress. Robinson et al., (2008) found that Fulton's body condition factor was positively associated with survival time during a thermal test, suggesting that physiological condition can influence how fish respond to elevated temperatures. Accordingly, thermal limits can directly impact the behavior, potential growth rate and distribution of fish.

A study in the United States investigated the upper thermal limits of brook trout in relation to their growth (Chackwick & McCormick, 2017). The results showed a decline in growth rate as the temperature exceeded 16°C, with growth ceasing at 23.5°C. Suggesting optimal growth of brook trout is between 13°C and 16°C (Chackwick & McCormick, 2017). Consequently, they argue that higher water temperatures directly can affect animal distribution (Chackwick & McCormick, 2017), suggesting that thermal increases will play a critical role in the future brook trout declines (Wenger et al., 2011). Diurnal thermal fluctuations with high daily maximum temperatures will also trigger long term cellular stress response (Chadwick & McCormick, 2017). Furthermore, temperature affects the competition between brook trout and brown trout. In Wisconsin, where brook trout are native and brown trout are invasive, the competitive suppression of brook trout by brown trout was reduced in cooler summer water temperatures (Olson et al., 2024), indicating brook trout has a competitive benefit in cooler waters. Generally, brook trout is more frequently detected in upper stretches of watercourses with colder water (Havs- och vattenmyndigheten, 2023).

1.4 Aim and hypothesis

The aim of my thesis is to compare the temperature preferences of both species and to discuss the significance of these behaviors for present and future distribution of invasive brook trout in Sweden. In addition, I will also observe if their temperature preference changes as the temperature increases during the day. Because thermal preference has not previously been compared between brown trout and brook trout populations in Sweden, my hypotheses are derived from studies on brook trout and brown trout behavior in their native ranges and in other regions. Ultimately, this study aims to fill this scientific gap by providing empirical data if the species' temperature preferences differ.

My hypotheses for this study were:

H_1: Brook trout will have a lower temperature preference than brown trout.

H_0: There is no difference in temperature preference between brook trout and brown trout.

2. Material and method

2.1 Study area, sample collection and ethical permits

The experiment was conducted in a facility belonging to Sportfiskarna Jönköping 40 minutes outside of Jönköping, Sweden in the spring 2026. The study species were brook trout (n=15) and brown trout (n=15) (fig. 1). The fish were around 1 year old and all individuals originating from the river Kallån in Ydre municipality. The experiment was done with the ethical and animal testing permits; Idnr 007328 and Dnr 5.8.18-04260/2026.

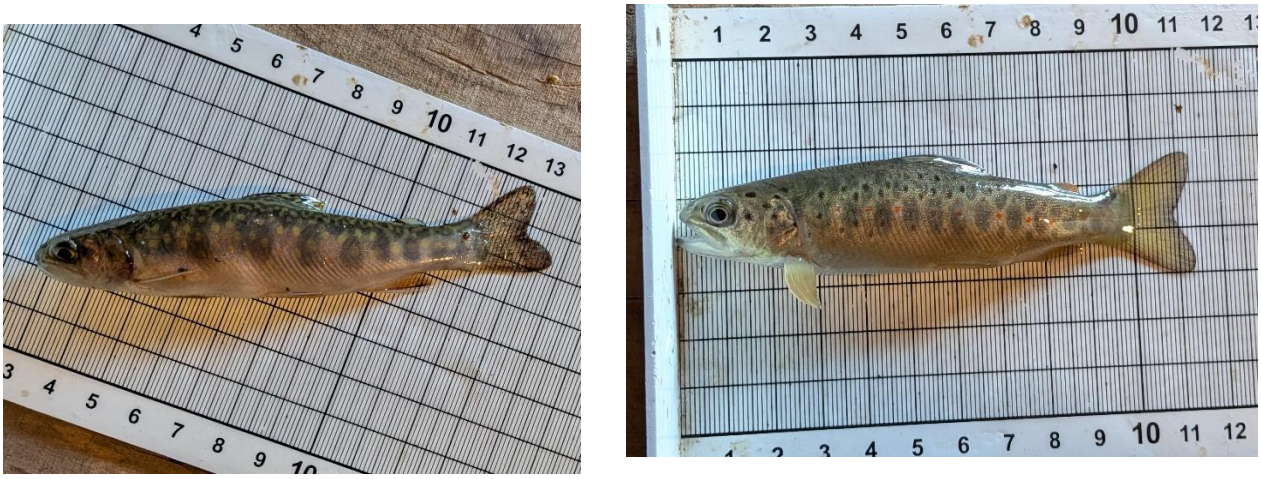


Figure 1. Brook Trout (Salvelinus fontinalis) (left), brown trout (Salmo trutta) (right). Photos by Sandra Andreasson.

2.2 Experiment setup

2.2.1 Temperature preference annuli

To calculate the temperature preference of the fish, a circular artificial tank, called a temperature preference annuli (TPA) was used (figure 2). This setup was first designed and tested by Myrick et al., (2004), the replicate used in this experiment was earlier build by Gräns et al., (2012). The TPA consisted of 3 channels and a middle part with a water outlet, these were separated by plexiglas. The outer channel worked as a mixing channel for the heated and non-heated water and the inner channel worked as an outlet channel that allowed water to end up in the middle and then discharge. The middle channel, formed between the outer and inner channel, served as a swimming channel for the fish participating in the experiment.

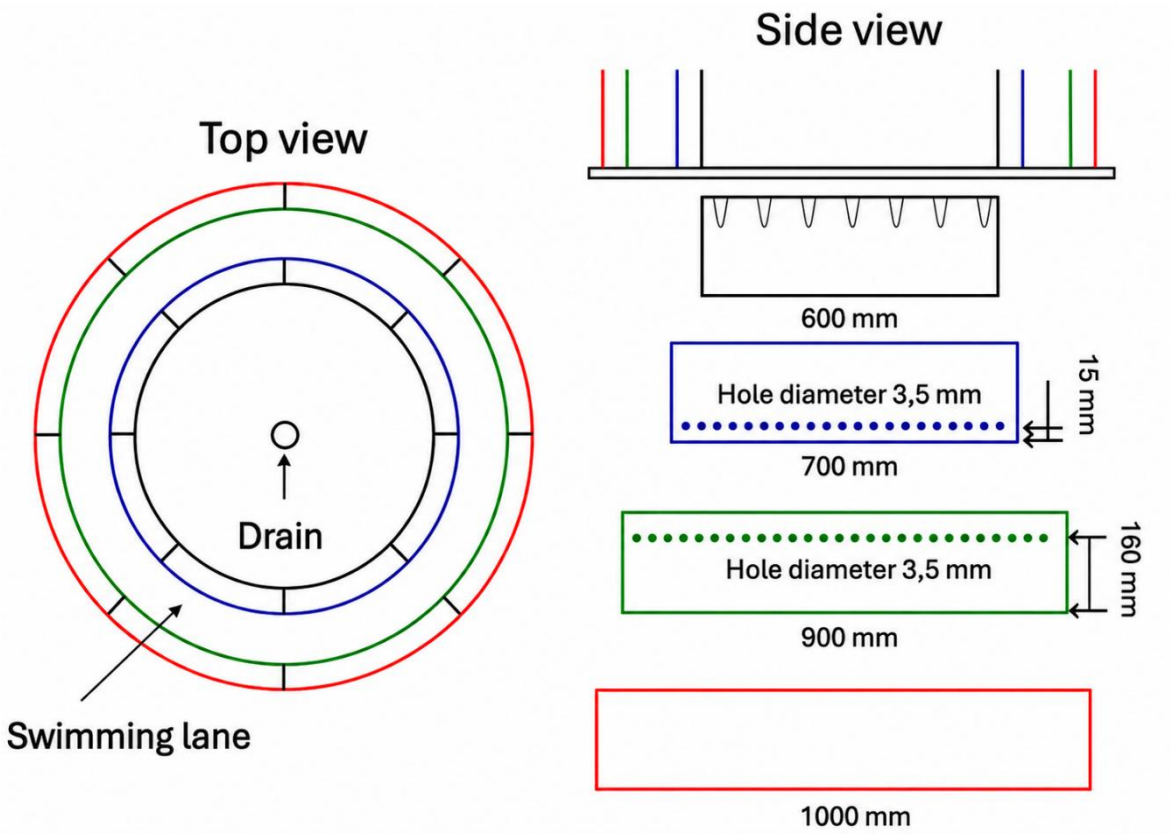


Figure 2. Diagram of the temperature preference annuli. Red circle = first (outer) disc, green circle = second disc, blue circle = third disc and black circle = fourth (innermost) disc. The red and green discs formed the outer channel (mixing channel). The green and blue discs formed the middle channel with a width of 10.5cm (swimming channel). The blue and black discs formed the inner channel (outlet channel). Outer and inner channels were divided into 8 parts placed with a 45° angle from both the middle and each other. The outermost circular disc was intact. All the sections in the outer channel were 34 cm long and 5,5 cm wide. In both the two next discs were small holes placed at the top edge and bottom edge, to enable the passage of water. The disc creating the inner wall of the outer channel had 32 holes per section and a diameter of 3,5 mm, placed at the top below the water's surface. The third creating the outer wall in the inner channel. The sections in this channel were 27,5 cm long and 5,5 cm wide and contained of 25 holes with a diameter of 3,5 mm placed in the bottom. The innermost disc had 8 recessed sections where the water fell over when the level exceeded approximately 17 cm, and had a diameter of 100 cm. In the center of the innermost disc, was an outlet with a diameter of 15 cm where the water was abducted from the TPA. The whole TPA had a circumference of 314,5 cm and a diameter of 15 cm. Marginally modified from a drawing by A.Gräns och M. Axelsson. Used with permission.

2.2.2 Materials

The incoming water came directly from a millpond with connection to the lake Assjön. The water was led to the TPA from two plastic containers (20L) with four tubes from each container with differential pressure, one with heated water and the other with unheated. The heated water was warmed up with four thermostatic heaters (from Eheim Jäger), 2 units at 75 watts and 2 units at 150 watts. Water went from there to the TPA with the help from four plastic tubes, with an inner diameter of 1 cm and a length of 205 cm, from each container. To reduce the flow, a cut-off pipette was placed into each tube to reduce the opening to 3 mm. The flow in the mixing channel amounted to 0,59 L/min, but this varied somewhat because of difficulties to regulate incoming valves and with that the volume in each plastic containers. The temperature of the incoming water varied throughout the day because the water from the millpond warmed up by the sun during the day, creating a difference in the temperature variance in both the mixing and swimming channel. The lowest temperature in the channel varied from 5.9 to 10.6°C and the highest temperature varied from 8 to 13.2°C. This resulted in a temperature gradient ranging from 5.9 to 10.3°C for

the coldest water and 8-13.2°C for the warmest, where the average temperature difference between warm and cold water was 2.3°C. The temperature was measured with 8 temperature sensors (type K), that was placed with a 45º gap along the inner wall in the middle channel. The temperature sensors were marked with 1-8. A PicoLog (USB TC-08, with 0.5°C accuracy) data logger was connected to the temperature sensors, with a continuous registration of the temperature via the software PicoLog 6.

To minimize disruptions of the fish the TPA's outside was covered with a 1 mm thick white paper. The illumination consisted of a light ramp (Growth Technology T5HO 4x24W, 240 V ~ 50 Hz, 96W – 0.4 Amp) placed above the TPA to minimize shadow formation into the swimming channel. The light was connected to a timer with a photoperiod of 12L/12M. A window was also covered to reduce the impact from extern light sources.

2.2.3 Experiment design and data collection

The tests were conducted over a period of 3 days from 21st to 23rd of April (9:00 am – 16:00 pm) alongside the electrofishing carried out by educated staff from Sportfiskarna Jönköping. The electrofishing was done prior to each trial day, giving the fish time over night to acclimatize to the new environment. Two 60-liters plastic boxes with drilled holes in the sides, were placed in a waterlogged pool (1x1m). The holes allowed water to flow through the boxes and the pool. One box was market for brook trout and the other for brown trout. The pool was constantly filled with water from the millpond, to ensure new incoming water. Stones were placed in the boxes as hiding places and to reduce stress. Before the tests, the fish were placed in the boxes, and afterwards, they were moved to the pool to separate them from the fish that had already gone through the experiment.

Each fish was individually placed in the swimming channel, and everyone was introduced at temperature sensor number four. The introductions alternated between brook trout and brown Trout. The fish had the opportunity throughout the whole experiment to move and change swimming direction. The acclimatization period was 10 minutes, and the test lasted for 20 minutes, making a total of 30 minutes. The fish's temperature preference was recorded using a camera (DJI Osmo Action 5 Pro), positioned approximately 1 meter above the TPA using a mount attached to the wall. Simultaneously the temperatures were recorded in PicoLog using the temperature sensors. Immediately after the test, the fish were measured in weight and length. After the three days of experiments, the brown trout was again released into the river Kallån, while the brook trout were euthanized due to them being invasive.

2.3 Data analysis and statistics

Video recordings of each fish were analyzed manually. Movement periods and stationary periods were documented. The fish had to be still for more than 10 seconds to be documented as a stationary period, to be able to define it as a temperature choice. The position of a stationary fish was determined based on the position of the temperature sensors (1-8) and if a fish positioned between two temperature sensors, both were documented. The duration of each movement or stationary period was also noted.

Fulton's body condition (K) of each fish was calculated based on weight and length measured during the experiment (K=1 optimal body condition, K>1 over nourishment, K<1

undernourishment) (see Appendix 1). Furthermore, the temperature in the TPA varied slightly during the 20-minute test for each individual fish. The temperature was recorded at approximately one-minute intervals via the Picolog software. However, the total number of temperatures recorded varied across tests. Therefore, a mean temperature was calculated to obtain a single value for each position of the temperature sensors (1-8) for each individual test in the TPA. Finally, all raw data was compiled for further processing in R-Studio (R Core team, 2024).

2.3.1 Weighted temperature index and confounding temperature variable

The water temperature in the TPA was regulated by the millpond which caused the water to become warmer during the day. This made the water conditions similar to the natural water they live in. Though, it created a confounding temperature variable where each fish encountered different water conditions during the test. Further, it resulted in an obstacle comparing the average temperature at each position for different individuals, depending on their start time in the experiment. To address this an index ranging from 0 to 1 (with 1 representing the warmest preference) was calculated to measure each individual's temperature preference. The temperature index was calculated for each position the fish choose to remain stationary, regarding the minimum and maximum temperatures for the time a fish was in the TPA. The individuals holding multiple positions were assigned an index for each one.

$$Temperature\ index = \frac{(Temperature\ choice\ - Temperature\ min)}{(Temperature\ \max - Temperature\ \min)}$$

To account for differences in the amount of time a fish spent at each position, an index time was calculated. Each position's temperature was weighted according to the proportion of the total observation period the fish spent at the position. Thus, positions where the fish spent more time contributed more strongly to the calculated index time. For example, if a fish spent 15 minutes at one location and 2 minutes at another, the temperature of the first location was given a greater weight in the calculation because the fish spent a larger proportion of the observed period there. An index time for each temperature index was calculated, accordingly for every stationary position. Lastly, the weighted temperature index was calculated, where the sum of the index time was divided with the total time (total stationary time for all positions). This made it possible to give each fish an index value from 0-1 and to directly compare their temperature preference. This approach eliminates temperature as a confounding variable, allowing for an independent comparison between brook trout and brown trout's temperature preferences regardless of their individual temperature gradient in the TPA.

$$Index\ time = Temperature\ index\ \times Position\ time$$

$$Weighted\ temperature\ index = \frac{\sum(Index\ time)}{Total\ time}$$

By calculating a weighted temperature index for each fish their preference could be compared (see Appendix 2). These calculations were done using R-studio. Also plots and statistical test were made by using R-studio. A boxplot was developed to visualize the temperature preference for both species, and a linear model was conducted to evaluate if there was a difference in weighted temperature index between the two species. In addition, the linear model also evaluated

the interactive effects of body condition and species on the weighted temperature index. In addition, an average temperature preference was calculated for each individual, based on the time spent at each position, in order to also visualize their preferred temperature in degrees Celsius (see Appendix 2). However, this calculation did not take the confounding temperature variable into account, unlike the weighted temperature index, and therefore couldn't be directly compared between individuals.

An analysis was also conducted to determine if the confounding temperature variable could have had an impact on the fish's temperature preference over the course of the day. The start time for each individual test was found in the Picolog data and was documented in an excel file (see Appendix 2). A line chart was done in R-studio to analyze if the start time had an impact on the weighted temperature index. The start time was converted into decimal hours to create a continuous linear scale for the statistical analysis. A linear regression was conducted to investigate if the correlation between weighted temperature index and start time was different between species.

3. Results

3.1 Comparing temperature preference between species

Brook trout tended to prefer the colder water which was reflected in a significant difference between the species in the weighted temperature index (p-value = 0.027) (fig. 3). The mean for brown trout was 0.67 and for brook trout 0.43.

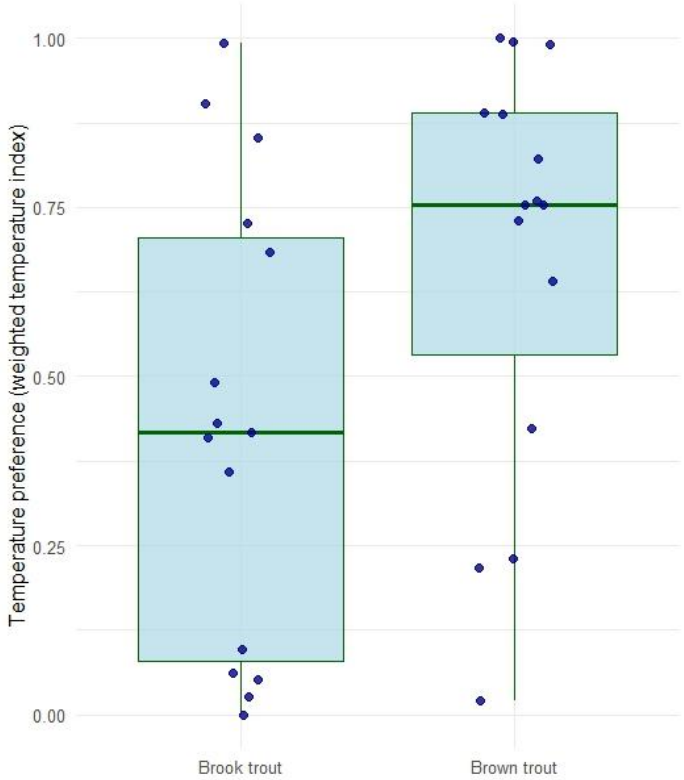


Figure 3. Boxplot of weighted temperature index mean for brown trout (mean = 0.67) and brook trout (mean = 0.43). Statistically significant difference was observed between the two species ($p = 0.027$, $R^2 = 0.26$).

3.2 Effects of body condition and species on weighted temperature index

The result shows that there is a statistically significant interaction between body condition and species ($p = 0.037$), indicating that the relationship between body condition and the weighted temperature index differs between the two species (fig. 4). For brown trout, body condition exhibits a negative relationship with the weighted temperature index ($\beta = -2.41$). In contrast, brook has a positive trend for the relationship between body condition and weighted temperature index ($\beta = 0.77$). A trend can be observed that the variation for brook trout is higher than for brown trout.

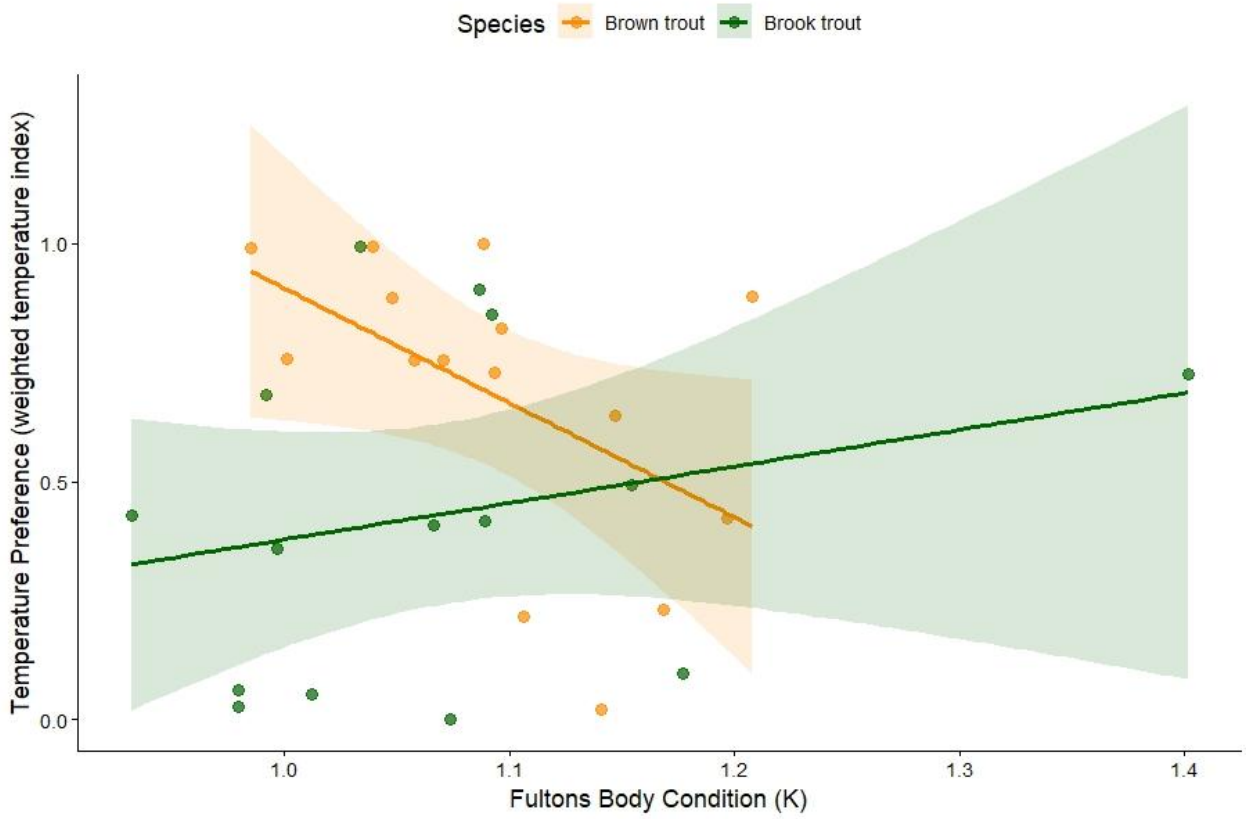


Figure 4. Relationship between body condition index and weighted temperature index for brown trout and brook trout. Points represent individual fish (n = 30). Lines show linear regression fits for each species derived from a general linear model, shaded areas representing 95% confidence intervals. A statistically significant interaction was detected ($p = 0.037$, $R^2 = 0.26$), showing a negative relationship for brown trout and a positive relationship for brook trout.

3.3 The confounding temperature variable's impact on temperature preference

There was no significant interaction between start time and species ($p>0.05$), indicating that the trend of the weighted temperature index over time didn't differ significantly between the two species (fig. 5). Visually a trend can be observed in fig. 5, where the brown trout's temperature preference is rather stable over time, unlike brook trout, where the trend appears to be more unstable and fluctuating.

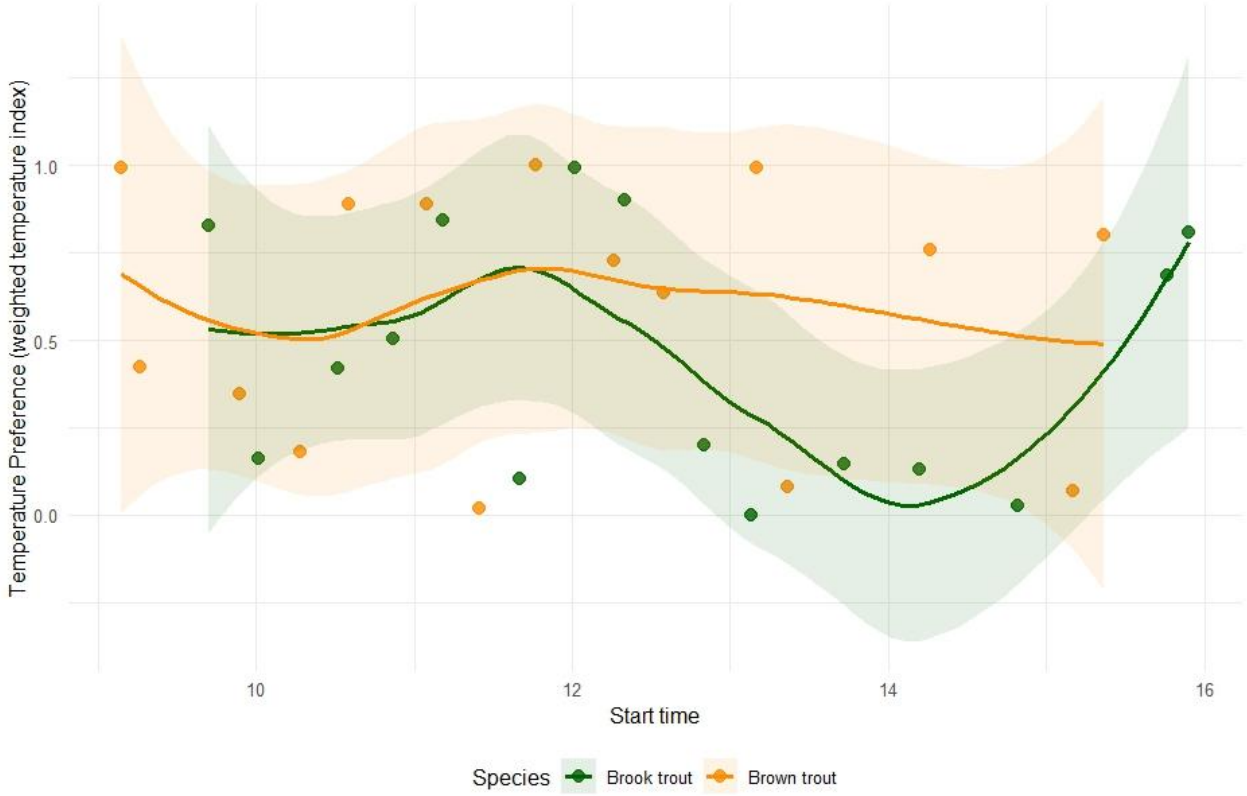


Figure 5. Diurnal variation in weighted temperature index for brook trout and brown trout. Points representing individual fish observation (n = 30). A non-statistically significant interaction was detected between weighted temperature index on start time between species (P >0.05).

4. Discussion

The result from my study indicated a difference in mean temperature preference between brown trout and brook trout, brook trout appeared to prefer colder water than brown trout when getting the opportunity to choose temperature freely. Furthermore, my results demonstrated that the effect of body condition on temperature preference differs between both species. Brown trout had a negative trend indicating that they tend to prefer higher temperatures as their body condition decreases, in contrast to brook trout that had a positive trend. In addition, the brook trout's temperature preference showed greater diurnal variation than brown trout. Overall, the results suggest that the temperature preferences were slightly more scattered for brook trout than for brown trout (fig. 3), making it difficult to draw firm conclusions. There is so far no research investigating the temperature preference of brook trout in Sweden, and therefore lesser understanding about its impact on distribution patterns, climate change and nature conservation involving methods to remove it.

4.1 Difference in temperature preference between species

The observed mean temperature preference was lower for brook trout than brown trout, this difference did reach statistical significance (p = 0.027). The trends in the experiments are interesting and could align with some other studies. For instance, it is shown that brook trout thrives better in colder water (Picard et al., 2003). Furthermore, in North America the suppression from brown trout is lower in colder waters (Olsen et al., 2024). This physiological temperature

behavior towards cooler water could provide a valuable predictive tool for management of brook trout outside of its native range, such as in Sweden. By understanding these thermal boundaries for the brook trout, it could help to predict current spatial distribution and identify cold water streams where brook trout are most likely to establish or persist. Furthermore, it is also shown that brook trout growth rate decreases when the temperature is 16°C or higher (Chadwick & McCormick, 2017). This provides an evident biological motivation for brook trout to actively select cooler thermal environments. In the present study, however, the maximum temperatures in the TPA did not reach this level. If a broader gradient with higher temperature had been used, a more pronounced behavioral response and clearer preference patterns could probably have been observed. However, even a narrow thermal gradient could be enough to get a response, as fish are directly affected by even small changes in temperature (Gräns et al., 2010).

When comparing the two species, brook trout showed substantial variation in both temperature preference and body condition, whereas brown trout exhibited much less variation. Brown trout showed a negative trend, where a body condition closer to optimal (K=1) corresponded to a higher weighted temperature index (temperature preference). This suggests that brown trout with optimal or even higher body condition (K≤1) may possess sufficient energy resources to actively seek out their preferred thermal conditions. Furthermore, individuals with a higher body condition factor might have enough energy reserves to temporarily tolerate conditions outside their thermal preference, as physiological conditions can influence the temperature response for fish (Robinson et al., 2008). However, the small temperature gradient in this study may have limited the ability to fully observe this behavior.

In contrast, the variation observed in brook trout regarding the effect of species and body condition on weighted temperature index, could be attributed to its invasive nature. Phenotypic plasticity in non-native aquatic species often provides a significant competitive advantage during establishment in new environments (Davidson et al., 2011). Introduced brook trout display high plasticity in foraging behavior and microhabitat selection, allowing them to exploit resources and outcompete native species (Zavorka et al., 2017). This adaptive capacity may explain the contrasting outcomes between the two species. The positive trend observed in brook trout suggests that individuals with higher body condition (K>1) prefer warmer temperature to accelerate digestion, consistent with them being ectothermic (Magnuson et al., 1997). Additionally, the pronounced variation in brook trout could come from higher individual-based variation, which is also linked to their high capacity for behavioral plasticity (Zavorka et al., 2017). Overall, this could explain the difference between the species' outcome in the experiment.

4.2 Thermal limits and climate change

It was expected that brook trout would have a lower thermal preference in the afternoon compared to the morning when the water temperature elevated. Although there was no statistical significance, a more distributed pattern could be observed for brook trout than brown trout over the course of the day (fig. 5). This could indicate stressed behavior caused by the thermal gradients. Selecting a cooler temperature during higher temperature peaks could mitigate stressed behavior and allow brook trout to prevent body temperature from reaching upper metabolic thresholds (Chadwick & McCormick, 2017). In streams where brook trout and brown trout live in sympatry, having the flexibility to adjust temperature selection diurnally could be particularly advantageous for brook trout. Previous research in United States, have shown that thermal refugia is vital for brook trout survival when living together with brown trout (Hitt et al., 2016). This implies that brook trout seeking cooler habitats during elevated temperatures possibly can avoid

high temperature competition (Olsen et al., 2024). This adaptation for brook trout relies on a stream with thermal heterogeneity. If cooler habitats are unavailable, brook trout may have a difficult time avoiding these thermal peaks and thus the competition pressure from brown trout will become higher. However, it's important to highlight that brown trout, as well as brook trout, could require colder habitats as temperatures increase.

Understanding these limits for brook trout is especially interesting in the context of climate change. My study was conducted in the middle of May, meaning the stream temperatures had not yet reached the highest temperatures typical of the summer months (June, July and August). Considering that our highest water temperature reached 13.2°C, it is likely that the water temperature in summer will reach the thermal limits for brook trout (Chadwick & McCormick, 2017). Though it must be noted that the water we used from the mill pond is stagnant and could heat up quicker than a stream. In Sweden, the effects of climate change have led to a decline in juvenile abundance of brown trout in the south while it is increasing in the north (Donadi et al., 2023). This could suggest a decline in brook trout populations in south of Sweden as well, as my results showed that they prefer colder water than brown trout.

4.3 Further research

Some experiment limitations must be considered when interpreting the results of this study. I would argue for one study being a relatively small sample size (n = 15 per species). A larger sample size would have made it possible for other statistical tests and stronger correlations. Furthermore, it was difficult to control the flow and regulate incoming water from the valves. Which can have had an impact on the condition of the fish in the TPA. When recording the films of each test, the camera did not have a perfectly right angle, which created a shadow from the discs in the TPA, where it was difficult to see the fish. In those scenarios I had to predict the spot, which was possible because the shadow only covered a small part. I would argue that my results, indicating a difference in temperature preference for brook trout and brown trout, could provide a further interest in conducting more research in this area. Future studies should incorporate a larger sample size to reduce the risk of random variation and a non-significant result. Additionally, extending the duration of each trial in the TPA should be considered to obtain more reliable data. This method has only been implemented by Sportfiskarna Jönköping once prior to this project, therefore this study function as a pilot-study that could help improve future experimental approach. Several new research questions were raised during this study, such as whether age, sex, life stage, or season could influence their thermal preference. It would also be valuable to investigate social dominance behavior between the two species, by placing both in the TPA to observe how interspecific interactions impact their preference. Further, to investigate their physiological stress response towards temperature changes. Finally, determining the optional exposure time in the TPA remains an important obstacle to being able to distinguish true thermal preference from exploratory behavior.

4.4 Conclusion

In conclusion, my results showed that brook trout exhibits a lower temperature preference within this experimental setup, offering valuable insights into their ecological distribution in natural water courses. Furthermore, the effect of body condition on weighted temperature index differed between the two species. These behavioral variations may be attributed to brook trout being an invasive species with higher phenotypic plasticity (Davidson et al., 2011; Zavorka et al., 2020). Overall, brook trout had a broader variation in temperature preference, which could suggest that

they have the capacity to adapt their strategies and behavior to new environments. Over the course of the day, a slight trend indicated that brook trout sought out lower temperatures as the water warmed, although high variation made it difficult to predict with certainty. Ultimately, my findings suggest a clear trend towards brook trout having a lower temperature preference compared to brown trout.

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References

Chadwick, J. G., & McCormick, S. D. (2017). Upper thermal limits of growth in brook trout and their relationship to stress physiology. *Journal of Experimental Biology.* 220, 3976-3987. doi:10.1242/jeb.161224

Cucherousset, J., Aymes, J. C., Poulet, N., Santoul, F., & Céréghino, R. (2008). Do native brown trout and non-native brook trout interact reproductively? *Naturwissenschaften*, *95*(7), 647–654. https://doi.org/10.1007/s00114-008-0370-3

Davidson, A. M., Jennions, M., & Nicotra, A. B. (2011). Do invasive species show higher phenotypic plasticity than native species and, if so, is it adaptive? A meta-analysis. *Ecol Lett*, *14*(4), 419–431. https://doi.org/10.1111/j.1461-0248.2011.01596.x

Donadi, S., & Näslund, J. (2023). *Contrasting long-term trends in juvenile abundance of a widespread cold-water salmonid along a latitudinal gradient: Effects of climate, stream size and migration strategy*. Dryad Digital Repository. https://doi.org/10.5061/dryad.5tb2rbp85, version: sha256:aba2110d3746676b8989d8f73b03329ff7e2e8c3711dfe3a025697336fe1e439.

Elliott, J., & Allonby, J. (2013). An experimental study of ontogenetic and seasonal changes in the temperature preferences of unfed and fed brown trout,Salmo trutta. *Freshwater Biology*, *58*, 1840–1848. https://doi.org/10.1111/fwb.12173

Ern, R., Andreassen, A., & Jutfelt, F. (2023). Physiological Mechanisms of Acute Upper Thermal Tolerance in Fish. *Physiology*, *38*. https://doi.org/10.1152/physiol.00027.2022

Grande, M., & Andersen, S. (1990). Effect of two temperature regimes from a deep and a surface water release on early development of salmonids. Regulated Rivers: Research & Management, 5, 355–360

Gräns, A., Olsson, C., Pitsillides, K., Nelson, H. E., Cech, J. J., Jr., & Axelsson, M. (2010). Effects of feeding on thermoregulatory behaviours and gut blood flow in white sturgeon (Acipenser transmontanus) using biotelemetry in combination with standard techniques. *J Exp Biol*, *213*(Pt 18), 3198–3206. https://doi.org/10.1242/jeb.043570

Gräns, A., Rosengren, M., Niklasson, L., & Axelsson, M. (2012). Behavioural fever boosts the inflammatory response in rainbow trout Oncorhynchus mykiss. *Journal of fish biology*, *81*, 1111–1117. https://doi.org/10.1111/j.1095-8649.2012.03333.x

Havs- och vattenmyndigheten. (2023). Bäckröding (Salvelinus fontinalis). Retrived 2026-08-28 from https://www.havochvatten.se/arter-och-livsmiljoer/invasiva-frammande-arter/sok-frammande-arter/fakta/backroding.html#h-Hurleverde

Hitt, N., Snook, E., & Massie, D. (2016). Brook trout use of thermal refugia and foraging habitat influenced by brown trout. *Canadian Journal of Fisheries and Aquatic Sciences*, *74*, 406–418. https://doi.org/10.1139/cjfas-2016-0255

Höjesjö, J., & Austad, B. (2026). *Bäckrödingen – effekter på ekosystemet och metoder för att ta bort den* (Report 7215). Naturvårdsverket. https://www.naturvardsverket.se/publikationer/7200/978-91-620-7215-5/

Ipbes. (2023). *Thematic Assessment Report on Invasive Alien Species and their Control of the Intergovernmental Science-Policy Platform on Biodiversity and Ecosystem Services*. IPBES Secretariat. Retrieved August 10, 2026 from https://doi.org/10.5281/zenodo.7430682

Jobling, M. (1981). Temperature tolerance and the final preferendum-rapid methods for the assessment of optimum growth temperatures. Journal of Fish Biology. https://doi.org/10.1111/J.1095-8649.1981.TB05847.X

Korsu, K., Huusko, A., & Muotka, T. (2007). Niche characteristics explain the reciprocal invasion success of stream salmonids in different continents. *Proceedings of the National Academy of Sciences*, *104*(23), 9725–9729. https://doi.org/10.1073/pnas.0610719104

Korsu, K., Huusko, A., & Muotka, T. (2009). Invasion of north European streams by brook trout: Hostile takeover or pre-adapted habitat niche segregation? *Biological Invasions*, *12*, 1363–1375. https://doi.org/10.1007/s10530-009-9553-x

Korsu, K., Heino, J., Huusko, A., & Muotka, T. (2012). Specific Niche Characteristics Facilitate the Invasion of an Alien Fish Invader in Boreal Streams. *International Journal of Ecology*, *10*. https://doi.org/10.1155/2012/813016

Lau, J., & Terhorst, C. (2015). Causes and consequences of failed adaptation to biological invasions: The role of ecological constraints. *Molecular ecology*, *24*(9), 1987-1998. https://doi.org/10.1111/mec.13084

MacCrimmon, H. R., & Campbell, J. S. (1969). World distribution of Brook Trout, Salvelinus fontinalis. *Journal of the Fisheries Research Board of Canada*, 26, 1699–1725.

Magnuson, J., Crowder, L., & Medvick, P. (1979). Temperature as an Ecological Resource. *American Zoologist*, *19*, 331–343. https://doi.org/10.1093/icb/19.1.331

Myrick, C., Folgner, D., & Cech, J. (2004). An Annular Chamber for Aquatic Animal Preference Studies. *Transactions of the American Fisheries Society*, *133*, 427–433. https://doi.org/10.1577/02-154

Naturvårdsverket. (2025). *Invasiva främmande arter: Definition*.Retrieved from 2026-08-10 from https://www.naturvardsverket.se/amnesomraden/invasiva-frammande-arter/vad-ar-ifa/definition/

Nyman, L. (n.d.). Bäckröding (*Salvelinus fontinalis*) [Illustration]. SLU Artdatabanken. https://artfakta.se/taxa/206232/information

Olson, K. W., Pechacek, K., & Benike, H. (2024). Brook Trout population response to Brown Trout removal by electrofishing in a Wisconsin Driftless Area stream. *North American Journal of Fisheries Management*, *44*(3), 735–744. https://doi.org/10.1002/nafm.11008

Picard, C., Bozek, M., & Momot, W. (2003). Effectiveness of Using Summer Thermal Indices to Classify and Protect Brook Trout Streams in Northern Ontario. *North American Journal of Fisheries Management*, *23*, 206–215. https://doi.org/10.1577/1548-8675(2003)023<0206:EOUSTI>2.0.CO;2

SLU Artdatabanken. (2026a). *Artfakta: bäckröding (Salvelinus fontinalis).* Retrived 2026-04-12 from https://artfakta.se/taxa/206232/information

SLU Artdatabanken. (2026b). *Artfakta: öring (Salmo trutta).* Retrived 2026-04-12 from https://artfakta.se/taxa/100127/information

Robinson, M. L., Gomez-Raya, L., Rauw, W. M., & Peacock, M. M. (2008). Fulton's body condition factor K correlates with survival time in a thermal challenge experiment in

juvenile Lahontan cutthroat trout (Oncorhynchus clarki henshawi). *Journal of Thermal Biology*, *33*(6), 363–368. https://doi.org/https://doi.org/10.1016/j.jtherbio.2008.05.004

Simon, K. S., & Townsend, C. R. (2003). Impacts of freshwater invaders at different levels of ecological organisation, with emphasis on salmonids and ecosystem consequences. *Freshwater Biology*, 48, 982–994.

Smith, D., Jackson, D., & Ridgway, M. (2019). Thermal habitat of Brook Trout in lakes of different size. *Freshwater Science*, *39*, 56–69. https://doi.org/10.1086/707488

Stewart, E. M. C., Middel, T. A., & Ridgway, M. S. (2026). Brook trout thermal habitat selection in stratified lakes: diel patterns, body size, and the importance of thermal time. *Canadian Journal of Zoology*, *104*, 1–18. https://doi.org/https://doi.org/10.1139/cjz-2025-0087

Vitousek, P. M., D'Antonio, C. M., Loope, L. L., Rejmánek, M., & Westbrooks, R. (1997). Introduced species: a significant component of human-caused global change. *21*(1), 1-16.

Wenger, S. J., Isaak, D. J., Luce, C. H., Neville, H. M., Fausch, K. D., Dunham, J. B., Dauwalter, D. C., Young, M. K., Elsner, M. M., Rieman, B. E., Hamlet, A. F., & Williams, J. E. (2011). Flow regime, temperature, and biotic interactions drive differential declines of trout species under climate change. *Proceedings of the National Academy of Sciences*, *108*(34), 14175–14180. https://doi.org/10.1073/pnas.1103097108

Závorka, L., Koeck, B., Cucherousset, J., Brijs, J., Näslund, J., Aldvén, D., Höjesjö, J., Fleming, I., & Johnsson, J. (2017). Co-existence with non-native brook trout breaks down the integration of phenotypic traits in brown trout parr. *Functional Ecology*, *31*, 1582–1591. https://doi.org/10.1111/1365-2435.12862

Závorka, L., Larranaga, N., Lovén Wallerius, M., Näslund, J., Koeck, B., Wengström, N., Cucherousset, J., & Johnsson, J. (2019). Within-stream phenotypic divergence in head shape of brown trout associated with invasive brook trout. *Biological Journal of the Linnean Society*, *129*, 347–355. https://doi.org/10.1093/biolinnean/blz192

Appendix 1: Weight, length and body condition

Fish	Species	Weight (g)	Length (mm)	Body condition
1	Brown trout	18.2	115	1.196679543
2	Brook trout	18.1	122	0.996779466
3	Brown trout	17.9	114	1.208199014
4	Brook trout	22.9	128	1.091957092
5	Brown trout	22.3	127	1.088663595
6	Brook trout	17.4	117	1.086404768
7	Brown trout	14.6	114	0.985458414
8	Brook trout	15.8	114	1.066454996
9	Brown trout	21	128	1.001358032
10	Brook trout	16.1	118	0.979895705
11	Brown trout	13.7	109	1.057891368
12	Brook trout	7.6	87	1.154132935
13	Brown trout	17.9	116	1.146777236
14	Brook trout	19.3	121	1.089434685
15	Brown trout	13.2	108	1.047858558
16	Brook trout	13.4	111	0.979796451
17	Brown trout	12.3	104	1.093465521
18	Brook trout	15	114	1.012457274
19	Brown trout	25.8	133	1.096641768
20	Brook trout	16.1	111	1.177218124
21	Brown trout	12.4	102	1.168479695
22	Brook trout	16.3	118	0.992068322
23	Brown trout	15	113	1.039575243
24	Brook trout	12.4	96	1.401548032
25	Brown trout	23.2	128	1.106262207
26	Brook trout	11.1	106	0.931977404
27	Brown trout	15.6	111	1.140658555
28	Brook trout	5.7	82	1.033792313
29	Brown trout	11.7	103	1.070715741
30	Brook trout	9.5	96	1.073766638

Appendix 2: Start time, average temperature preference and weighted temperature index

Day (1-3)	Fish	Species	Start time	Average temperature preference (C°)	Weighted temperature index (0-1)
1	1	Brown trout	9:16 am	6.78	0.42
1	2	Brook trout	10:01 am	7.34	0.36
1	3	Brown trout	10:35 am	8.56	0.89
1	4	Brook trout	11:11 am	8.94	0.85
1	5	Brown trout	11:46 am	9.31	1
1	6	Brook trout	12:20 am	9.45	0.9
1	7	Brown trout	13:10 pm	10.41	0.99
1	8	Brook trout	13:43 pm	9.60	0.41
1	9	Brown trout	14:16 pm	10.63	0.76
1	10	Brook trout	14:49 pm	9.75	0.03
1	11	Brown trout	15:22 pm	11.54	0.75
1	12	Brook trout	15:54 pm	11.39	0.49
2	13	Brown trout	9:54 am	9.26	0.64
2	14	Brook trout	10:31 am	9.01	0.42
2	15	Brown trout	11:05 am	10.3	0.89
2	16	Brook trout	11:40 am	9.56	0.06
2	17	Brown trout	12:16 pm	10.68	0.73
2	18	Brook trout	12:50 pm	9.55	0.05
2	19	Brown trout	13:22 pm	11.94	0.82
2	20	Brook trout	14:12 pm	10.32	0.1
2	21	Brown trout	15:10 pm	10.87	0.23
2	22	Brook trout	15:46 pm	12.18	0.68
3	23	Brown trout	9:09 am	9.25	0.99
3	24	Brook trout	9:42 am	8.80	0.73
3	25	Brown trout	10:17 am	8.58	0.22
3	26	Brook trout	10:52 am	8.98	0.43
3	27	Brown trout	11:25 am	9.06	0.02
3	28	Brook trout	12:01 pm	10.25	0.99
3	29	Brown trout	12:35 pm	10.55	0.75
3	30	Brook trout	13:08 pm	9.43	0