Deciphering lifelong thermal niche using high-resolution otolith δ18O thermometry exemplified within supplemented lake trout (Salvelinus namaycush) populations

Olivier Morissette1,3*, Louis Bernatchez1, Michael Wiendenbeck2, Pascal Sirois3

1 Institut de Biologie Intégrative des Systèmes (IBIS), Université Laval, Quebec City, Québec, Canada
2 German Research Centre for Geosciences (GFZ), Potsdam, Germany
3 Chaire de recherche sur les espèces aquatiques exploitées, Département des sciences fondamentales, Université du Québec à Chicoutimi, Chicoutimi, Québec, Canada

* Corresponding author

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ABSTRACT

1. The selection of water temperature habitat by fish is strongly regulated by physiology and behaviour. However, delineation of a species lifelong thermal niche remains technically challenging. Lake trout (Salvelinus namaycush) survival and productivity are recognised as being tightly linked to a somewhat restricted thermal habitat. The factors guiding temperature selection during each life stage remain poorly understood.

2. In this study, we tested the significant factors controlling the realised thermal niche of lake trout from two southern Quebec small boreal lakes that experienced supplementation stocking during the last 20 years. We used oxygen stable isotope (δ18O) thermometry of otolith calcium carbonates using secondary-ion mass spectrometry (SIMS) to estimate experienced lifelong temperatures. We investigated the thermal habitat of lake trout with known genotypes (local, hybrid and stocked).

3. Ontogeny and genetic origin influenced temperature selection in both studied lake trout populations. Young-of-the-year consistently used warmer, shallower habitats (10.7 ± 3°C, 7.5 m depth) prior to a juvenile transition to colder and deeper waters (8.5 ± 3.3°C, 10 m depth). Stocked lake trout, originating from a genetically distinct ecotype, exhibited a more variable thermal niche, with some individuals consistently using warmer habitat (10.4 ± 1°C) than local fish. Their hybrid progeny also occupied a warmer thermal niche, intermediate to the parental strains. We propose that increased fat content and genetic origin are potential explanatory factors for warmer temperature use.

4. This study reiterates that high-resolution otolith δ18O thermometry is a uniquely well-suited approach for unravelling the multiple factors that influence lifelong temperature selection in fish.
Our results illustrate that the realised thermal niche is influenced by a genetic-environment interaction.
1 Introduction

Water temperature is a critical property in aquatic environments as it defines suitable habitats for ectotherms, such as fishes and amphibians. Thermal conditions can have a strong impact on both physiological (e.g., growth rates, reproductive success) and ecological processes (e.g., prey and habitat selection) (Clarke, 2006). There is an apparent correlation between fish physiology and adapted behaviour, whereby temperature preference and the performance of physiological functions co-vary for a given species or a specific life stage (Coutant, 1987). This suggests that fish actively seek a temperature that will maximise their fitness. Potential competition and partition of thermal habitats suggest that temperature should be envisioned as an ecological resource, comparable to trophic and reproductive resources (Magnuson, Crowder & Medvick, 1979). Indeed, the range of used temperatures could be considered as one of the n-dimensional niche axes according to Hutchinson’s (1957) definition of an ecological niche. However, a distinction should be made between the “realised thermal niche” and the preferred temperature, the latter only applying to temperature ranges measured in laboratory, under conditions where all other parameters are controlled (Bergstedt et al., 2012). A gap may then exist between temperature preference and occupancy, owing to complex ecological constraints imposed by the natural environment.

Quantification of the realised thermal niche of fishes remains technically challenging in natural environments. Over decades, the use of acoustic telemetry and internal archival tags have become the gold standard for in situ assessment of thermal niches (Bergstedt et al., 2003; Plumb & Blanchfield, 2009). However, fish capture and the surgical implantation of such tags can be time consuming and is generally unsuitable for small/young fish (Elrod, Ogorman & Schneider, 1996; Lucas & Baras, 2000). This limitation is also accompanied with the risk of low recapture rate of
tagged fish. Oxygen isotope ratios (δ¹⁸O) in biogenic carbonates (i.e., molluscan shells, foraminifera) can provide a reliable alternative method for *a posteriori* temperature estimations in fish (Urey *et al.*, 1951), when applied to otoliths material (Devereux, 1967; Darnaude *et al.*, 2014; Patterson, Smith & Lohmann, 1993). Otolith formation is the result of the progressive precipitation of biogenic aragonite (or vaterite in a smaller proportion) under conditions where the oxygen isotope ratio will be near equilibrium with ambient water via a temperature-dependent hydrolysis reaction (Campana, 1999; Høie, Otterlei & Folkvord, 2004). Quantifying whole-otolith δ¹⁸O values provides temperature information integrated across a fish’s entire lifetime, although it does not provide information regarding distinct periods of an individual fish’s life (Kalish, 1991). High-resolution transects of oxygen isotope ratios can be obtained by extraction of otolith material using micromilling, giving access to the isotopic ratios recorded within the daily pattern of otolith precipitation as well as their extension to individual bands that represent months to years (Campana & Thorrold, 2001; Pannella, 1971), providing a record of the lifelong *in situ* thermal niche across all life stages (Hanson, Wurster & Todd, 2010; Wurster, Patterson & Cheatham, 1999).

Subsampling otolith bands by micromilling is done using fine bit to drill into targeted regions of the otolith. The material removed is subsequently collected and analyzed using isotopic ratio mass spectrometry. However, micromilling is limited by the size of the otolith and annuli widths, which can prevent study of particular species or life stages. More recently, technical advances in ion beam sampling technologies coupled with high-precision, high-sensitivity isotope ratio mass spectrometry have opened the opportunity to quantifying oxygen stable isotope ratios using individual point-based analyses of otolith (Matta *et al.*, 2013). In particular, secondary-ion mass spectrometry (SIMS) offers highly precise stable isotope quantification. Both micromilling and SIMS provide comparable δ¹⁸O results, whereby the latter offers a higher spatial resolution.
(Hanson, Wurster & Todd, 2010). Previous research showed that otolith thermometry is uniquely suited for assessing the role of temperature as an ecological resource subject to competitive interactions and ontogenetic variations (Patterson, Smith & Lohmann, 1993; Kalish, 1991).

Despite growing access to these analytical techniques, otolith thermometry has been seldom used for thermal niche assessment of exploited fish species, especially at or before their sub-adult life stages (Shirai et al., 2018).

Predatory cold-water fish occupying deep temperate lakes are likely to exhibit competitive interactions for thermal habitats, as the volume of suitable habitat can be limited during the summer season (Christie & Regier, 1988; Murdoch & Power, 2013) when these lakes undergo thermal stratification (Wetzel, 2001). Lake trout (Salvelinus namaycush), a cold-water stenotherm living in deep oligotrophic lakes across North America, represents a very relevant fish model for exploring the importance of thermal resources (Martin & Olver, 1980). This species has a restricted optimal temperature range (8 – 12 °C, Coutant, 1987; Mackenzie-Grieve & Post, 2006; Plumb & Blanchfield, 2009) and a low tolerance for dissolved oxygen less than 6–7 mg/L (Evans, 2007; Evans, Casselman & Wilcox, 1991). These environmental preferences are likely to promote competition for thermal resources, which can be influenced by the ecological particularities of a studied population. There is a growing body of evidence that boreal and arctic salmonids show intraspecific variation in performance among ecotypes and strains in relation to temperature (Bergstedt et al., 2012; Bergstedt et al., 2003; McDermid et al., 2013). Accordingly, thermal niche use (based on otolith δ¹⁸O thermometry) was shown to differ between European whitefish (Coregonus lavaretus) morphs (Kahilainen et al., 2014; Kelly, Amundsen & Power, 2015), a difference mostly attributed to trophic niche use (benthic/pelagic preys). McDermid et al. (2013) demonstrated that small-bodied lake trout ecotypes usually seek out lower temperatures than large-
bodied ecotypes. Intra-specific variation of phenotypic traits influencing depth (and temperature) has also been documented. For instance, the ecotypic differences in depth and thermal preference have been linked to fat content (Eshenroder et al., 1995) and swim bladder gas retention, both determining the neutral buoyancy depth. Selective breeding of these ecotypes showed that both traits were heritable, with inter-ecotype hybrids displaying fat content and gas retention intermediate to parental values (Eschmeyer & Phillips, 1965; Ihssen & Tait, 1974). Those observations suggest that depth and thermal niche use are, at least partially, genetically determined and could be modified by inter-ecotype hybridization (Bergstedt et al., 2003).

Although the thermal niche of adult lake trout has been studied extensively, thermal habitat use of young-of-year (YOY) and juveniles is not well known. Temperature selection in lake trout is believed to be correlated with age, with younger fish preferring higher temperatures than adults (Bergstedt et al., 2003). Peck (1982) observed that lake trout YOY in Presque Isle Harbor, Lake Huron, exhibit a prolonged residence in relatively shallow waters (< 8 m) near spawning sites as long as water temperature does not exceed 15°C over an extended period. Likewise, young lake trout in Great Bear Lake, NWT, Canada concentrate in shallow shoreline waters from the ages of 0–3 years (Miller & Kennedy, 1948). Great Bear Lake juveniles and adults were largely overlapping in term of depth and thermal niche, probably linked to the absence of thermocline and/or predation gradient (Chavarie et al., 2019). Arctic charr (Salvelinus alpinus) and European whitefish were also shown by otolith δ¹⁸O thermometry to use warmer thermal niche, linked to use of shallower littoral zone in their first years of life, (Kahilainen et al., 2014; Murdoch & Power, 2013; Godiksen et al., 2012). Bronte et al. (1995) showed that lake trout fry in western Lake Superior are most abundant in shallow waters (10–15 m) in July, after which they migrate to deeper waters as they grow, being most abundant at 40–49 m depth in October. In contrast, other studies
have suggested that YOY lake trout seek deeper and colder habitats shortly after yolk-sac absorption (Martin, 1951; Deroche, 1969; Royce, 1951). Precise factors influencing YOY and juveniles thermal habitats remain to be identified.

Despite the majority of lake trout populations existing in small lakes (Gunn & Pitblado, 2004), data on thermal habitat use is mostly coming from larger systems. Inter-population particularities, exemplified by ecotypes identification, could have significant impacts on thermal niche. Two common ecotypes (e.g., planktivorous and piscivorous) are recognized in small boreal lakes, their expression reflecting the combined influence of environmental conditions, food availability (e.g. access to pelagic prey fish) and genetics (Bernatchez et al., 2016; McDermid, Shuter & Lester, 2010). The planktivorous ecotype is characterized by a low growth rate, early maturation (~ 6 years) and a shorter maximum length of fish (< 450 mm) and is associated with lakes where large pelagic preys (cold-water pelagic fish) are absent. Piscivorous ecotype, which is feeding on large pelagic preys, exhibit high growth rates, late maturation (> 9 years) and a larger (> 600 mm) maximum length (Bernatchez et al., 2016; Houde & Scrosati, 2003). Few lakes host both ecotypes in sympatry, the vast majority only hosting a single allopatric ecotype.

This study aimed to document the relative role of different biotic (e.g. life stage, genetic origin, trophic position) factors driving thermal habitat use by lake trout. We assessed lifelong thermal niche use through high spatial resolution SIMS otolith oxygen stable isotope (δ\(^{18}\)O) thermometry. The resulting high-resolution temporal estimations of experienced temperatures are used to describe the thermal habitat of YOY and juvenile fish and shed light on the uncertainties surrounding early life thermal habitats of lake trout. Our study targeted fish having distinct genetic origins (i.e., local, stocked or hybrid, see method section) from two allopatric lake trout populations of small-bodied planktivorous ecotype. Both lakes had been previously stocked for angling
supplementation with source populations of a large-bodied piscivorous ecotype (Morissette et al., 2018). By combining genotype-by-sequencing technologies and SIMS stable isotope measurements, we tested whether fish ontogeny and genetics were significant factors influencing temperature selection by lake trout. We hypothesised that stocked and hybrid trout would tend to use warmer habitats than local fish due to their genetic background (piscivorous ecotype), which is shown to be more related to warmer thermal niche (McDermid et al., 2013).

2 Methods

2.1 Study systems and supplementation stocking history

Stocking has been used for lake trout population supplementation for over a century (Kerr & Lasenby, 2001). In Quebec, Canada, 46% of lakes hosting lake trout angling-exploited population have been stocked at least once since 1928 (Ministère du Développement Durable de l’Environnement de la Faune et des Parcs, 2013). In most cases, captive-reared first-generation progeny of wild breeders captured from allopatic lake trout populations of the piscivorous ecotype were stocked, no matter the ecotype of recipient populations. Breeders were captured from known spawning sites in source lakes for eggs and milt collection. Eggs were artificially fertilized in hatcheries and progeny reared in captivity until stocking. Neither domesticated strains nor adult fish have ever been used for the stocking of these lakes (Morissette et al., 2018).

2.2 Fish sampling and processing

We sampled two lakes, one in 2012 (Lake Louisa; 45.769°N, 74.419°W) and 2013 (Lake McFee; 45.714°N, 75.623°W) using the same experimental fishing protocol. Both lakes are small (<500 ha) and deep (Z_max >56 m), located in southern Quebec, Canada, and both display similar summer thermal stratification (Figure 1). Both lakes host an allopatic population of a
planktivorous ecotype (small-bodied) lake trout and both lack large pelagic prey, such as pelagic forage fish (i.e. *Coregonus* spp. or *Osmerus mordax*) or large invertebrates (*Mysids* spp.). These two fish communities are comprised mostly of white sucker (*Catostomus commersonii*), yellow perch (*Perca flavescens*), pumpkinseed (*Lepomis gibbosus*), rock bass (*Ambloplites rupestris*) and small cyprinids (Figure 1). Both lakes were stocked multiple times from the same source population (Blue Sea Lake, Quebec). The most recent stocking event in both lakes took place no more than 12 years before our sampling. These lakes were characterised previously as displaying a relatively balanced proportion of purely local and stocked fish as well as their hybrids (Valiquette *et al.*, 2014; Morissette *et al.*, 2018).

We collected 75 fish per lake using the gill net method from a normalised lake trout sampling protocol, in collaboration with the Québec Ministère des Forêts, de la Faune et des Parcs (Service de la faune aquatique, 2011). For each fish, we measured total length (TL, mm) and mass (g) in the field. The adipose fin was sectioned and stored in 95% ethanol in individual plastic vials (Eppendorf, Mississauga, Ontario). For both lakes at their deepest point, we collected temperature profiles at each 1-meter depth up to 20 meters and surface and thermocline water samples using clean Niskin bottles. Water samples were transferred in the field to airtight, nitric acid–washed (HNO₃, trace metal grade) Nalgene bottles and conserved at room temperature prior to δ¹⁸O analyses. In the laboratory both of the sagittal otoliths were extracted from the lake trout using nitric acid–washed plastic forceps. Otoliths were washed with ultrapure water, dried and stored in Eppendorf plastic vials before further processing.
2.3 Genetic assignment of individual fish

Each fish was assigned a genetic origin (i.e., local, stocked or hybrid) based on their single nucleotide polymorphism (SNP) genotype using next-generation sequencing (genotype-by-sequencing; GBS) and the software ADMIXTURE (Alexander, November & Lange, 2009). Our methodology is presented in detail in Morissette et al. (2018). Briefly, to assign the fish genotype we used the individual admixture proportions ($Q$), defined as the proportion of subsample parts of the genome most likely to be related to the stocking source (Blue Sea Lake) compared to the local (wild) genotype. We classified fish as being stocked when $Q_{stocking \ source} + SE \geq 90\%$, local fish when $Q_{stocking \ source} + SE \leq 10\%$, and hybrid when $Q$ values fell between 10 and 90%. No further distinction was attempted between hybrid classes because the number of fish analyzed here would have been too small (5 hybrids per lake, see below) in each category to draw any rigorous interpretations.

2.4 Preparation of otoliths and ageing of fish

The right sagittal otolith was embedded in a two-component epoxy resin (Miapoxy 100, Freeman, OH, USA) and cut into 1-mm-thick traversal sections using a slow-speed diamond-bladed saw (IsoMet saw; Buehler, IL, USA). After sectioning, we amplified the annuli contrasts by progressive grinding and polishing with sandpaper (2000 grit Wetordry™, 3M™) and aluminium oxide lapping film (1- and 5-μm lapping film, 3M™). Periodical observations of the grinding stage were realised during polishing, which guided the grinding/polishing procedure. Digital images of each otolith were captured using a digital camera (Leica DMC) coupled to a dissection microscope (Leica MZ12) at a 30 – 60x magnification. Age counts and increment measurements (μm) were done from the nucleus to the maximum ventral radius of the otolith (longest axis) following established methods and criteria (Casselman & Gunn, 1992; Simard & Magnin, 1972) using ImageJ v 10.2
software (Abramoff, Magalhaes & Ram, 2004). Two independent readings (two readers) were done and a third additional count/measurement if the first counts were not in agreement (~25% of otoliths). Using estimated age of the fish and length at capture, we back-calculated length-at-age (for complete method, see Morissette et al., 2018), using the body-proportional hypothesis (Francis, 1990). We selected a subset of 30 fish of age >10 years (mean = 13.7 ± 2.4 years) based on the lake and genetic origin (2 lakes × 3 genetic origin [local, hybrid or stocked] × 5 fish = 30 total).

2.5 SIMS otolith δ¹⁸O values analytical methods

Two SIMS mounts were required to accommodate all 30 otolith samples while ensuring that all areas to be measured remained within an 8-mm radius from the center of the circular, 25.4-mm diameter mount. Each mount contained 20 crystals of IAEA-603 and three (3) sub-millimetre pieces of UWC-3 calcite reference materials (IAEA, 2016; Kozdon et al., 2009). To minimise the amount of epoxy in each block—thereby minimising the amount of sample outgassing within the SIMS ultra-high vacuum system—polycarbonate discs were milled to provide individual wells for each otolith as well as a separate well for receiving the two calcite reference materials. We verified surface topography using white light interferometry, and we found the topography to be <5 microns for all regions of interest.

The samples were cleaned in an ultrasonic bath of high-purity ethanol before being carbon sputter-coated for subsequent scanning electron microscopy (SEM) imaging. We produced both secondary electron and backscattered electron images for each otolith to obtain information regarding the locations of cracks within the material and also to provide clear identification of the main growth axis of a given otolith. After completing the initial SEM imaging, an additional 35-nm thick, high-
purity gold film was sputter-deposited directly on top of the carbon coating. Both mounts were then imaged fully using the stitching software of a Nikon Eclipse motorised optical microscope. These images were then loaded into the SIMS point logger software.

We used the Cameca 1280-HR secondary-ion mass spectrometer of the German Research Centre for Geosciences (Potsdam) to produce $\delta^{18}$O profiles. SIMS $\delta^{18}$O analyses employed a ca. 2 nA mass filter $^{133}$Cs$^+$ beam having a Gaussian density distribution. The beam was focused to a ca. 5 µm diameter on the polished sample surface. The total impact energy of the Cs$^+$ ions was 20 keV. Each analysis was preceded by 25 × 25 µm rastered pre-sputter for 80 s. Charge compensation was achieved using low-energy, normal incidence electron flooding. To suppress within-run drift of the isotope ratio, a 15 × 15 µm raster was used during data collection, thus ensuring a flat-bottom crater geometry. The dynamic transfer option of the secondary column ion optics was used to compensate for the rastering of the primary beam during data collection. Analyses were performed as point profiles from the otolith core and following the longest axis with stepping distances of ca. 100 µm with both reference materials being analysed (typically after every 10th acquisition); the sequence of data acquisition was not randomised.

Secondary ions were accelerated by a -10 kV potential applied to the non-magnetic, stainless sample holder. Before the start of data acquisition, the SIMS conducted automated centering routines for the field aperture in both the X and Y directions and the contrast aperture in the X direction only. The instrument had a field of view of 80 × 80 µm, in conjunction with a 40 V energy band pass. The mass spectrometer was operated in a static multi-collection mode with the $^{16}$O$^-$ being collected in the L2’ Faraday cup and the $^{18}$O$^-$ signal being collected in the H2’ cup; the amplifier system used thermally stabilised $10^{10}$ Ω and $10^{11}$ Ω amplifiers. Magnetic field drift of the mass spectrometer was eliminated effectively by the use of an NMR-controlled feedback loop. The
mass resolution of the instrument was determined to be \( M/dM \approx 1,800 \), which is effectively the full
transmission of the 1280-HR and is fully sufficient to eliminate both the \(^{16}\text{O}^{1}\text{H}_2\) and \(^{16}\text{O}^2\text{H}\)
isobaric interferences from the \(^{18}\text{O}\) mass station. A single analysis consisted of 20 integrations of
4 s each, resulting in a data collection time of 80 s per analysis (equivalent to a total of \( \sim 3 \) min per
point when including the sample moving, pre-sputtering and automatic centering routines). This
analytical design resulted in a count rate of \( ca. 2.8 \times 10^9 \) ion/sec on the \(^{16}\text{O}\) mass station. A total of
375 \( \delta^{18}\text{O} \) analyses were conducted on the 30 otoliths, along with 46 determinations on both the
IAEA-603 and UWC-3 calcite reference materials. All analyses were conducted over three days
and required a total of 31 hours of data acquisition. A 3sd filter was applied to all data with respect
to the 20 individual integrations conducted during the given analytical run.

To quantify the instrumental mass fractionation and to monitor the possible occurrence of a time-
dependent drift during our runs over the hour-long period, we used two calibration materials. We
detected a drift equivalent to 0.098‰ per hour toward higher \(^{18}\text{O}/^{16}\text{O}^-\) values as defined by \( n = 9 \)
determinations of the UWCV-3 calcite. This drift occurred solely on the third day of data
acquisition. Therefore, we applied a corresponding linear drift correction to all data collected on
this specific day. To calibrate the instrumental mass fractionation of our SIMS instrument on each
separate day, we used the value of \( \delta^{18}\text{O}_{\text{VPDB}} = -2.37 \) for the IAEA-603 (IAEA, 2016), and we used
the \( \delta^{18}\text{O}_{\text{VSMOW}} = 12.49 \) for the UWC-3 (Kozdon \textit{et al.}, 2009). For converting between the two
oxygen isotope scales, we used the equation on page 440 of Brand \textit{et al.} (2014). All analytical
results are reported in standard \( \delta \) notation (‰) relative to Vienna Standard Mean Ocean Water
(VSMOW).
2.6 Quantification of the water $\delta^{18}$O isotopes

The $\delta^{18}$O values of surface and thermocline water samples were analysed at the Ján Veizer Stable Isotope Laboratory (University of Ottawa, Canada). The $\delta^{18}$O$_{\text{water}}$ values were determined using a Finnigan MAT Delta plus XP + Gasbench; a precise water volume (0.2 mL) was pipetted into an Exetainer vial. The vials were flushed and filled off-line with a gas mixture of 2% CO$_2$ in helium. The flushed vials were left at room temperature for a minimum of 24 h. The CO$_2$ gas was analysed automatically under continuous flow during this period. The results were normalised to Vienna Standard Mean Ocean Water (VSMOW) using three calibrated internal standards that spanned most of the natural range. The routine precision (2σ) of the analysis was ±0.15‰. Lake-specific $\delta^{18}$O$_{\text{water}}$ values were assumed to represent the average of the surface and thermocline results.

2.7 Calculation of experienced temperatures

Interspecific difference in life history and physiological mechanisms could influence oxygen isotope fractionation in otoliths (Høie, Otterlei & Folkvord, 2004; Weidman & Millner, 2000). Hence, many authors advise caution on the use of universal (e.g., multi-specific) temperature-mediated $\delta^{18}$O fractionation equation (Hanson, Wurster & Todd, 2010; Rowell et al., 2005). Hence, species-specific fractionation equation (or developed for closely related species) should be preferred (Murdoch & Power, 2013; Storm-Suke et al., 2007). Accordingly, we estimated temperatures derived from the otoliths $\delta^{18}$O using an oxygen isotope fractionation equation developed for Salvelinus species, in a similar geographic distribution that of our study (Storm-Suke et al., 2007); this equation has been shown to produce reliable temperature estimation for charr species. Lake-specific $\delta^{18}$O$_{\text{water}}$ values were used to calculate experienced temperature ($T^\circ{\text{C}}$). The equation is expressed as the difference between otolith ($\delta^{18}$O$_{\text{otolith}}$) and water ($\delta^{18}$O$_{\text{water}}$) values:
\[ \delta^{18}\text{O}_{\text{otolith}} - \delta^{18}\text{O}_{\text{water}} = 32.90 - 0.23 \times T^\circ C \]

We transformed spot analyses positions (X, Y) along each otolith data acquisition axis of the SIMS analysis to fish age and length using the digitally measured position of each annulus along the data acquisition axis. We then used a non-linear regression of annuli position as a function of age to estimate the age of the specific fish at every point of analysis. Age estimates were then used for estimation of TL (mm) using an individual-specific von Bertalanffy growth model (VBGM). Length-age and age-annulus models were fitted using the FSA package in the R statistical software (R Core Team, 2016). The growth model fitting is presented in detail in Morissette et al. (2018).

2.8 Statistical analyses

We modelled the effects of age and genetic origins on the occupied thermal habitats (response variable temperature) of lake trout YOY using a linear mixed-effect model. Linear mixed model was fitted using the function lme in the R package nlme (Pinheiro et al., 2019). The factors of the model were genetic origin (fixed, two levels: local and hybrid) and age (fixed, two levels: hatching and end of first growing season). Individual fish nested within a lake were treated in the model as a random slope to allow for the experimental repeated-measure design and individual variability. We did not include an estimated temperature of the two first growing seasons for stocked fish in our evaluation. Stocked fish $\delta^{18}$O_{otolith} values were representative of the hatchery habitat (mean = $19.07 \pm 1.22\%_{\text{VSMOW}}$) rather than that of the sampled lakes (mean = $22.89 \pm 0.88\%_{\text{VSMOW}}$); therefore, the estimated temperatures were not biologically relevant ($> 20^\circ C$, max = $38.4^\circ C$). This difference was only observed in the first two growth seasons, which is in agreement with the age at stocking of hatchery fish (1+).
We used a similar model to establish the thermal habitats used by sub-adult fish (< 6 y) using a new factor for age (fixed, six levels: 1 to 6). We ran pairwise comparisons between age classes using the lsmeans function (lsmeans package) in R (Lenth, 2016). Since this analysis showed that the realised thermal habitat stabilised after the fourth growth season, we modelled the influence of genetic origin (fixed, three levels: local, hybrid and stocked) and age (continuous variable) on post-settlement thermal habitat (>fourth growth season) with individuals nested within the lake as a random slope.

To test for the biotic factors influencing the temperature used by adults, we modeled the effects of ecological individual variables on temperature at capture (estimated temperature at the last annuli). Biotic variables included in the model were total length (TL, mm), percentage of fish population attributed to stocked genotype (Q\text{stocked}), C:N ratio (a measure of fat content), trophic position (TP) and Δ\text{Pelagic} (contribution of pelagic prey to diet); C:N, TP and Δ\text{Pelagic} were estimated from stable isotope (δ\text{13}C and δ\text{15}N) from white muscle collected on the same fish (see Morissette et al. (2019) for complete methodology). We tested the effects of biotic variables (TL, Q\text{stocked}, C:N, TP and Δ\text{Pelagic}) on temperature at capture using a similar linear mixed-effect model with lake as a random factor.

3 Results

Selected fish from both lakes were similar in terms of age (mean = 13.7 ± 2.4 years) and total length (mean = 451 ± 82 mm TL). However, some stocked fish exhibited a strikingly larger size than observed for the local or hybrid fish (Table 1). Total observed length difference was consistent with our recent observation that ~20% of stocked lake trout in allopatric populations of the planktivorous ecotype had a significantly larger size compared to local and hybrids (Morissette et al.)
These stocked fish had a body length more typical of lake trout of the piscivorous
type (i.e., their genetic origin). Otolith radius (μm) were found to correlate linearly with fish
TL (oto\textsubscript{radius} = 2.4 \times TL + 398.4, p < 0.05, R^2 = 0.76), where the largest fish have an otolith radius
~ 500 μm longer than smaller fish.

### 3.1 SIMS otolith δ\textsuperscript{18}O

For the three days of data collection, we recorded 1s repeatability for the IAEA-603 material of ± 0.30‰ (n = 16), ± 0.94‰ (n = 22), and ± 0.30‰ (n = 8), respectively. For the concurrent UWC-3 calcite, we recorded 1s repeatability for the three days of data acquisition of ± 0.11‰ (n = 15), ± 0.16‰ (n = 23), and ± 0.17‰ (n = 9). We found that the IAEA-603 calcite contained significant isotopic heterogeneity at the sub-nanogram sample mass provided by our SIMS instrument. This is consistent with the results reported by Nishida and Ishimura (2017) who observed variations in the isotopic compositions of IAEA-603 at the single-grain scale.

On the second day of our SIMS analytical sequence, we observed a clear relationship between the measured \(^{18}\text{O}/^{16}\text{O}\) ratio and the \(^{16}\text{O}\) count rate with some 2.5‰ difference in the isotopic ratio detected between the two groups of the IAEA-603 grain types (opaque and translucent). It is noteworthy that Nishida and Ishimura (2017) found a δ\textsuperscript{18}O value difference of only 0.28‰ between the two subpopulations they observed for IAEA-603. However, these authors were working with test portion masses some four orders of magnitude larger than masses used in our SIMS determinations. As suggested by Nishida and Ishimura (2017), the isotope ratio for individual grains of IAEA-603 could be correlated with the optical clarity of a given grain.

Based on the above observations, we based our otolith data reduction solely on the results from the UWC-3 calibration material. The observed repeatability of UWC-3 indicated that the overall
uncertainty on the individual otolith results was less than ± 0.2‰ (1s). Calculating the mean \(^{18}O^{16}O\) ratios from the multiple grains analysed from IAEA-603, we obtained \(\delta^{18}O_{VPDB}\) values of -2.29‰, -2.86‰ and -2.36‰ for each of the three analytical sessions, respectively. These results are in reasonable agreement with the assigned value for IAEA-603 of \(\delta^{18}O_{VPDB} = -2.37 \pm 0.04\‰\) (IAEA, 2016). We therefore believe that our absolute results are at least accurate to 0.5‰, whereas the relative difference between any two analytical results is reliable at the 0.3‰ (1s) level or better. Hence, these results highlight the need for caution regarding the possibility of calibration-related issues when working at a high resolution and with such a small sample mass.

We measured an average of 12 SIMS analyses per otolith (stepping distance 99.6 μm/step) distributed evenly over the entire length of a given otolith. No analysis failed to provide a result. Two spots were systematically done within the first annuli (first growing season) for every fish. Furthermore, a single point of analysis for the subsequent growing seasons was successful until the sixth growing season, where points of analysis had an above-annual frequency since annuli were narrower than our ~100 μm step distance. As expected, the temporal resolution provided by this methodology is significantly higher than that achieved by micromilling (e.g. 76 – 450 μm/spot; Hanson, Wurster & Todd, 2010). The median observed otolith \(\delta^{18}O_{VPDB}\) value was -7.60‰ (\(\delta^{18}O_{VSMOW} = 23.08\‰\)) with values ranging from -13.45‰ to -5.76‰ (\(\delta^{18}O_{VSMOW} = 16.74\‰\) to 24.98‰). There was no significant difference between surface and thermocline water \(\delta^{18}O_{VSMOW}\) values, but a significant difference was observed between our two lakes (Lake Louisa = - 8.05 ± 0.04‰, Lake McFee = - 7.32 ± 0.26‰).
3.2 Thermal habitats of young-of-the-year and juvenile lake trout

Yearlings form both lakes occupied a wide spectrum of thermal habitats, ranging from 5.82°C (5th percentile) to 15.52°C (95th percentile). The linear mixed-effects model revealed no significant effect of age (hatching versus the end of the first growth season) or genetic origin (local and hybrid) on the estimated used temperatures (Table 2) in the first year of life. The average estimated temperature at hatching was 10.88 ± 2.95°C (standard error, SE) and yearlings lived in a relatively constant thermal habitat during the entire first growing season. However, we observed pronounced inter-individual variability in terms of thermal habitat being occupied. Temperature at hatching was a significant predictor of the magnitude of temperature change during the first growing season ($\Delta T = -0.87 \times T_{\text{hatching}} + 9.45$, $p = 0.002$, $R^2_{\text{adj}} = 0.40$, Figure 2). Lake trout hatching at 10.9°C were the least likely to initiate a change of temperature; other individuals born in warmer or colder waters migrated to reach a thermal habitat closer to the observed temperature preference (10.9°C).

The temperature range occupied by juvenile lake trout was greater than that occupied by either adult or YOY fish, ranging between 3.45°C and 18.60°C, which is within the range of observed temperatures based on our determined depth-temperature profiles (Figure 1). The temperature range occupied by juveniles was influenced significantly by age; there was a significant difference between occupied temperatures after the third growing season (Table 3). Our model showed a significant ontogenetic effect on thermal habitats with older (> 3 years old) lake trout inhabiting colder temperatures (3°C colder) than younger juvenile and YOY (Figure 3). There was no significant effect of genetic origin on the thermal habitat being used. Back-calculated length-at-age from individual Von Bertalanffy models showed that movements to colder water was initiated at a TL of 100 mm and completed at ca. 250 mm TL (Figure S1).
3.3  Adult thermal habitats

The estimated habitat temperatures for adult fish ranged from 2.61°C to 16.95°C (mean = 8.32 ± 2.90°C). These values are all within the assumed species’ boundaries of thermal habitat (Plumb & Blanchfield, 2009). Modelling of adults’ thermal habitats showed a significant effect of genetic origin; stocked fish were using significantly warmer waters than local and hybrid fish (Table 4). However, lifelong experienced temperature at the adult life stage of stocked lake trout showed important between- and within-individual variability (Figure 4). At some point during their lives several fish occupied warmer habitats (average = 10.39°C) than the remainder of the stocked (average = 8.33°C), local and hybrid lake trout (average = 7.76°C). Two stocked fish from Lake McFee experienced a clear shift in their thermal niche at ca. 400 mm TL (Figure 4, Figure S2), a consistent behaviour exhibited for subsequent years.

Model of temperature at capture showed that Q_{stocked} and C:N (fat content) both had a significant positive relationship on temperature at capture (Table 5). According to this model, both fat content and Q_{stocked} were factors promoting use of warmer temperature (Figure 5). Interestingly, hybrid fish exhibited the use of thermal habitat intermediate to that of their parents, the hybrids more closely related to stocked genetic origin (high Q_{stocked}) displaying use of warmer temperature, comparatively to hybrid related to the local genotype (low Q_{stocked}), using colder water temperatures at the time of capture (Figure 5, bottom panel).
Discussion

High-resolution analyses of oxygen stable isotopes provided life-long estimation of lake trout thermal habitat. This study showed a marked difference between YOY/juvenile and adult thermal habitat. Later in life, inter-individual difference in thermal habitat emerged, potentially linked to genetic origin and related phenotypic traits. It has long been presumed that lake trout possess a very narrow thermal optimum, a perception largely influenced by Christie and Regier (1988) who showed a strong positive correlation between lake trout angling yield and the volume of lake that encompassed the 8–12°C thermal habitat. Following this pioneering work, the assessment of lake trout thermal habitat became of increasing interest for wildlife management and ecological theory. Thus, a variety of techniques were deployed to quantify the temperature range of the species. Notably, intensive gillnet sampling surveys (Elrod, Ogorman & Schneider, 1996; Elrod & Schneider, 1987), bottom and otter trawls (Peck, 1982), archival tags (Bergstedt et al., 2012; Bergstedt et al., 2016), acoustic and radio telemetry (Mackenzie-Grieve & Post, 2006; Plumb & Blanchfield, 2009) and remotely operated vehicles (Davis, Carl & Evans, 1997) were used to document trout habitat use in terms of depth and temperature. All of these methods proved to be useful, but they remained time- and resource-intensive and were limited by the size of fish, biased toward fish of larger size. From those studies, few studies reported on the thermal habitat of YOY and juvenile lake trout in natural settings (Bronte et al., 1995; Miller & Kennedy, 1948; Peck, 1982). However, Landsman et al. (2017) showed that otolith δ¹⁸O thermometry could provide an estimate of lake trout natal thermal habitat (lake Michigan) which were consistent with field observations (Bronte et al., 1995). Accordingly, our study is reiterating that δ¹⁸O thermometry based on the high-resolution SIMS analysis of otoliths is a powerful method for assessing the thermal niche of any size class of fish.
4.1 Assessment of the young-of-the-year thermal habitats

Yearlings consistently occupy waters having an average temperature of 10.7 ± 3.0°C. Our results match the preferred temperatures of lake trout YOY observed under controlled laboratory settings: 11.7°C (McCauley & Tait, 1970) and 9.0–11.5°C (Peterson, Sutterlin & Metcalfe, 1979). During their first growth season, individual lake trout displayed a high variability in the occupied temperature, but they tended to seek out a specific range of temperatures, initiating movements to avoid temperatures >14°C and <8°C, as suggested in the literature (Bronte et al., 1995; Peck, 1982). This temperature range corresponded to the upper metalimnion (depth = 7.5 m) in the sampled lakes during the period of summer thermal stratification (Figure 1). As such, our results support the hypothesis of Peck (1982) of extended residency in shallow (<8 m) waters.

However, the inter-individual variability we observed provides insight into the lack of consensus in the literature around thermal habitats during the early life stages of lake trout. Contrasting YOY behaviour could be linked to the different thermal conditions prevailing in lakes inhabited by lake trout, influencing early life tactics. Here, several individuals made marked migrations between thermal habitats (>6°C) during their first growing season. Yearlings tended to “escape” stressful thermal situations, whereas those hatching in warmer sites modified their position to reach colder habitats closer to their thermal optimum (and vice-versa). This is consistent with the results observed for populations inhabiting lakes experiencing rapid warming in the spring (i.e., southern or small lakes) in which YOY exhibit a rapid migration to lakes’ deepest parts (Martin, 1951; Royce, 1951). Accordingly, littoral residency has been observed in northern (or large) lakes where littoral zones, for most of the growing season, displayed water temperatures less than 16°C, such as in Great Bear Lake which does not experience any summer stratification (Miller & Kennedy, 1948; Johnson, 1975) and Lake Superior (Bronte et al., 1995; Peck, 1982).
Estimated temperatures from otolith cores show that local fish hatched under more variable temperatures compared to hybrids, suggesting a broader use of spawning sites for local fish compared to hybrids (Figure 2). Hence, while wild lake trout exhibit a certain degree of spawning site fidelity (Binder et al., 2015), hatchery-reared fish may not have the ability to recognize high-quality spawning sites (Bronte et al., 2007) or may even reproduce at a different time (Krueger, Swanson & Selgeby, 1986). This hypothesis remains to be tested. Admittedly, our interpretation of otolith core data should be taken with caution. Indeed, material accumulated in otolith core could be linked to mother-specific isotopic values, biasing the estimated temperature. However, there are still large uncertainties in maternally derived isotopes in otolith and no strong evidence exist in the presence of mother’s δ¹⁸O signature (Elsdon et al., 2008; Hegg, Kennedy & Chittaro, 2018). Caution is also warranted as no otoliths roasting was done before SIMS analyses, which would have suppressed organic matters present within the otolith structure. Intrinsic otolith organic material has been shown to potentially affect oxygen isotopic ratios (Matta et al., 2013), which would impact temperature estimations. Otolith primordium has previously been identified as organic fraction-rich zone (Jolivet et al., 2008), which can introduce additional variability of temperature estimation. However, observations of estimated temperature in the range of the species’ preference provide us confidence in our results.

Identifying a single thermal habitat in the same growing season may be obscured by seasonal variability in lake temperature. However, it should be noted that both sampled lakes are deep dimictic lakes that experience two turnover events (in spring and fall), and both have a relatively stable thermal stratification during the summer. Hence, according to the assumed seasonal shifts in thermal stratification, from the time of the eggs’ hatching (April) to the end of the growing season (October), the thermal zone with a temperature of ca. 10°C could be variable in term of its depth,
but would be available throughout this period, (Wetzel, 2001). In August-September, this thermal zone corresponds to a depth of 10 m (Lake Louisa) and 8.5 m (Lake McFee) (Figure 1). Our data could not assess the YOY use of horizontal microhabitats during the first growth season. However, they show clearly that these YOY used a relatively homogeneous thermal habitat, which was consistently warmer than that used by juvenile and adult conspecifics.

4.2 The onset of an ontogenetic thermal migration

The first significant changes in thermal habitat use were observed beginning in the third growing season, and this eventually stabilised within an “adult thermal habitat” during the fourth season. Juvenile lake trout displayed an increasing use of colder habitats: 1.5 ± 0.7°C and 3.2 ± 0.7°C colder than the thermal habitats occupied at the time of hatching for the third and fourth seasons, respectively. We suggest that predation pressure could be a factor contributing to this phenomenon. As many other top-predator fish, lake trout seek larger prey as they get older and grow (Martin, 1970; Pazzia et al., 2002; Sherwood et al., 2002). This ontogenetic diet shift is hypothesised to be the sum of the increasing capacity for eating larger prey (gape limit) and the need to achieve a greater energetic input (Mittelbach & Persson, 1998; Pazzia et al., 2002). Juvenile diet shifts in lake trout initiate normally ca. 150–200 mm TL, a size covering the 1- to 4-year age group (Martin, 1951; Trippel & Beamish, 1989). Accordingly, our length-at-age back-calculations had an average TL of 166.2 mm (± 20.6 SD) and 207.7 mm (± 23.9 SD), in their third and fourth growing seasons, respectively. Adult planktivorous lake trout feed extensively on large zooplanktonic prey, benthic macro-invertebrates and small littoral forage fish (Martin, 1966; Vander Zanden & Rasmussen, 1999).
The use of relatively warm, shallower habitats could be a juvenile life history tactic to avoid potential predation by older and larger conspecifics. Large lake trout living in a habitat lacking energy-rich pelagic prey items are prone to opportunistic feeding, including cannibalistic behaviour on smaller conspecifics (Morissette et al., 2018; Searle, Verde & Belk, 2018). As they grow, juvenile lake trout will eventually exceed the upper gape limit of the majority of adult predator taxa in the lake. Then, occupying the same thermal habitat as larger adult lake trout becomes less risky. Accordingly, based on equations published by Keeley & Grant (2001), the average adult lake trout in our study (mean = 451 mm TL) are not likely to feed on fish prey larger than 109 mm TL. This observation is consistent with our results, whereby the first evidence of thermal transition occurred ca. 100 mm TL. Therefore, when most of the juvenile fish settle into colder adult thermal habitat, their average total length (207 ± 24 mm TL) exceeds the size at which they are generally preyed upon—except for predation by lake trout that are > 800 mm TL. In summary, relatively warm, shallow habitats offer reasonable protection from potential predation by adult conspecifics as most, but not all (next section), adult fish rarely used thermal habitat > 10°C (Figure 4).

4.3 Adult temperature use: evidence for genetic × environment interactions?

The obtained δ¹⁸O values estimate a thermal niche for adult lake trout of 8.5 ± 3.3°C. This is consistent with the known adult lake trout thermal preference for 8°C (Bergstedt et al., 2003; Mackenzie-Grieve & Post, 2006). We observed that most fish rarely used thermal habitats > 12°C, the suggested upper limit of lake trout thermal preference (Plumb & Blanchfield, 2009), and lake trout avoided temperatures > 16°C entirely (Guzzo, Blanchfield & Rennie, 2017), the assumed upper threshold of lake trout’s thermal habitat (Guzzo & Blanchfield, 2016; Cline, Bennington & Kitchell, 2013; Plumb & Blanchfield, 2009). Lake trout from the stocked genetic group exhibited a more variable thermal niche, with some of the individuals experiencing long-term use of warmer
water temperatures of 12–16°C. However, it is unlikely that genetics alone explain thermal habitat use, as only a fraction of stocked fish exhibited this “warm water” behaviour. Our results tend to suggest that individual variation of thermal habitat of adult lake trout can be linked to the combined interaction of genetic and dietary elements.

In this study, those adult fish that consistently occupied a warmer thermal niche (12°C) were similar in terms of three conditions; a large size (> 450 mm TL), high fat content (C:N ratio) and genotype closely related to the source of stocking population (Figure 5). Exceptional growth within lakes hosting planktivorous populations could be promoted by a cannibalistic diet, targeting juvenile trout when other pelagic forage fish were absent, for which genetic origin (i.e. piscivorous ecotype) is probably a predisposition factor (Morissette et al., 2018). A realised thermal habitat of 10.3 ± 2.4°C for largest stocked fish is consistent with a thermal niche overlap with YOY lake trout (10.7 ± 2.9°C). The higher metabolic cost of long-term residency in warmer water could be mitigated by access to readily available, high-fat content prey. Feeding-motivated thermal excursions to warmer habitats are a common behaviour in some lake trout populations (Plumb & Blanchfield, 2009; Sellers et al., 1998; Guzzo, Blanchfield & Rennie, 2017; Morbey et al., 2006). High lipid content in lake trout, as well as being linked with high-fat content diet, may increase neutral buoyancy which in turn may facilitate vertical migration and affect depth use (Eschmeyer & Phillips, 1965; Eshenroder et al., 1995). Mc Dermid et al. (2013) showed that populations of large-bodied lake trout ecotypes were more tolerant to warm water and exhibited higher within-population variability in terms of thermal habitat. Tolerance of warmer environmental niches could therefore provide additional opportunities for larger stocked fish (Pépino, Goyer & Magnan, 2015), giving access to juvenile lake trout resources and other fish species present in the lake. Finally, the relationship observed between thermal habitat and stocked origin admixture proportions (Qstocked) in hybrid lake
trout (Figure 5) suggests that other factors, potentially genetically-determined, may influence thermal habitat use. Assessment of those factors is, however, beyond the scope of this work.

To conclude, our study reiterates the value of otolith biogenic carbonate thermometry based on $\delta^{18}O$ from high-resolution SIMS analyses as a tool for assessing lifelong temperature histories. This methodology can greatly increase the resolution and number of spots analyses by otolith compared to both whole-otolith analyses and micromilling. Estimated temperatures, for all life stages, were consistent with published boundaries for this species’ thermal habitat, providing high confidence in the accuracy of this technique. Our results support the recent and growing realization of a wider lake trout thermal niche than had been previously assumed (Challice, Milne & Ridgway, 2019; Plumb & Blanchfield, 2009). Many authors emphasised the importance of among-population variability in strategies for using available thermal resources in salmonids (Elrod, Ogorman & Schneider, 1996; McDermid et al., 2013; Bergstedt et al., 2012). Assuming a strict narrow thermal habitat is probably a simplistic view of the life cycle for species, such as lake trout, that exhibit high inter-population variability (Muir et al., 2015). By being influenced by both heritable and potentially adaptive phenotypic traits specific to local lake trout morphotype and ecotype (Bergstedt et al., 2003; Eshenroder et al., 1995), we suggest that thermal habitat use (and/or its associated traits) could be a component of the species’ local adaptation (Mackenzie-Grieve & Post, 2006; McDermid, Shuter & Lester, 2010). Our results support this view; genetically and phenotypically divergent fish (e.g. stocked from an exogenous population) exhibit an atypical use of warmer, shallower water as compared to the indigenous population. A significant effect was also observed on hybrid individuals, suggesting a genetically-based component. We stress that those observations are not a rigorous test of local adaptation, which require explicit assessment of fitness. Nevertheless, our observations are of particular interest given that small boreal lakes harbour the
vast majority of exploited and supplemented lake trout populations (Gunn & Pitblado, 2004). Admittedly, it remains to be determined whether our observations can be generalised to other populations. Improved understanding of the relationship between temperature use and intraspecific interactions will assist wildlife managers in improving and refining management and conservation practices.

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Data availability
All raw data and R scripts are available on an online Dryad repository (link will be provided upon acceptance)

References


Guzzo M.M. & Blanchfield P.J. (2016) Climate change alters the quantity and phenology of habitat for lake trout (Salvelinus namaycush) in small Boreal Shield lakes. Canadian Journal of Fisheries and Aquatic Sciences, 14, 871-884.


Muir A.M., Hansen M.J., Bronte C.R. & Krueger C.C. (2015) If Arctic charr *Salvelinus alpinus* is 'the most diverse vertebrate', what is the lake charr *Salvelinus namaycush*? *Fish and Fisheries, 17*, 1194-1207.


R Core Team. (2016) R: A language and environment for statistical computing. R foundation for
statistical computing, Vienna, Austria.

flow to nursery habitats of the Gulf corvina (Cynoscion othonopterus) Canadian Journal
of Fisheries and Aquatic Sciences, 62, 2874-2885.


bimodal size structure in a top predator. The Open Fish Science Journal, 11, 36-45.

(Salvelinus namaycush) in small Canadian Shield lakes with respect to temperature, dissolved oxygen, and light. Canadian Journal of Fisheries and Aquatic Sciences, 55, 170-179.

ichtyologique en eaux intérieures, Tome I, Acquisition de données. p. 137. Ministère des
Ressources Naturelles et de la Faune, Québec.

 enzymatic evidence for the energetic advantage of switching diet in wild-living fish. 
Canadian Journal of Fisheries and Aquatic Sciences, 59, 229-241.

Shirai K., Otake T., Amano Y., Kuroki M., Ushikubo T., Kita N.T., Murayama M., Tsukamoto K.

Simard A. & Magnin E. (1972) Méthode de détermination de l'âge et de la croissance du touladi,
Salvelinus namaycush Walbaum, du lac l'Assomption et du lac Tremblant, Québec. Le
Naturaliste Canadien, 99, 561-578.

fractionation equation for Salvelinus species. Rapid Communications in Mass
Spectrometry, 21, 4109-4116.

predicted from Cisco (Coregonus artedii) population structure and conductivity. Canadian
Journal of Fisheries and Aquatic Sciences, 46, 1531-1538.

paleotemperatures and temperatures of the upper cretaceous of England, Denmark, and the


Table 1. Average total length (mm) and age (year) and standard deviation (SD) for sampled lake trout. Lake trout are classified according to their genetic origins within lakes Louisa and McFee.

<table>
<thead>
<tr>
<th>Lake</th>
<th>Genetic origins</th>
<th>Sample (n)</th>
<th>Total length (mm)</th>
<th>SD</th>
<th>Age (year)</th>
<th>SD</th>
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<td>Louisa</td>
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<td>50</td>
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<td></td>
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<td>1.4</td>
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<td>412</td>
<td>30</td>
<td>13.6</td>
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<tr>
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<td>534</td>
<td>125</td>
<td>16.2</td>
<td>2.2</td>
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Table 2. Linear mixed-effect models of the response variables for the occupied temperature of the first growing season. Columns present the estimated differences (positive or negative) of group response variables with the model intercept, standard error (SE), degrees of freedom (df) and p-values of the factor. p-values in bold indicate significant differences (p < 0.05). The random parts show the number of tested groups and experimental units (Ngrp) and total observations.

<table>
<thead>
<tr>
<th>Coefficient</th>
<th>Response</th>
<th>Occupied temperature (°C) during the first growing season</th>
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<tr>
<td></td>
<td>Estimate</td>
<td>SE</td>
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<tr>
<td>Fixed parts</td>
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<tr>
<td>Intercept</td>
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<tr>
<td>Genetic origin (local)</td>
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<tr>
<td>Interaction (origin × age)</td>
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<td>2.65</td>
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</table>

<table>
<thead>
<tr>
<th>Random parts</th>
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<tr>
<td>Ngrp</td>
<td>15 fish in 2 lakes</td>
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<tr>
<td>Observations</td>
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Table 3. Linear mixed-effect model of juvenile (< 6 years) occupied temperature (response variable). Columns present the estimated differences (positive or negative) of factor response variables with the model intercept, standard error (SE), degrees of freedom (df) and p-values for the factor. p-values in bold indicate significant differences (p < 0.05). The random parts show the number of tested groups and experimental units (Ngrp) and total observations.

<table>
<thead>
<tr>
<th>Coefficient</th>
<th>Response</th>
<th>Juvenile occupied temperature (°C)</th>
<th>Estimate</th>
<th>SE</th>
<th>df</th>
<th>p-value</th>
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<td></td>
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<td></td>
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<tr>
<td>Intercept</td>
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<td></td>
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<tr>
<td>Growing season 3</td>
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<tr>
<td>Growing season 4</td>
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<td>-3.22</td>
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<td>Growing season 5</td>
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<tr>
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<td>20 fish in 2 lakes</td>
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<tr>
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Table 4. Linear mixed-effect model of temperature occupied by adults (response variable). Columns present the estimated differences (positive or negative) of factor response variables with the model intercept, standard error (SE), degrees of freedom (df) and $p$-values for the factor. $p$-values in bold indicate significant differences ($p < 0.05$). The random parts show the number of tested groups and experimental units ($N_{grp}$) and total observations.

<table>
<thead>
<tr>
<th>Coefficient</th>
<th>Response</th>
<th>Estimate</th>
<th>SE</th>
<th>df</th>
<th>$p$-value</th>
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<td>Adult occupied temperature</td>
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<td></td>
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<tr>
<td>Fixed parts</td>
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</tr>
<tr>
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<td>0.02</td>
</tr>
<tr>
<td>Genetic origin (local)</td>
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<td>-1.79</td>
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<tr>
<td>Ontogeny (age)</td>
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<td>0.07</td>
<td>163</td>
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<tr>
<td>Random parts</td>
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<tr>
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<td>30 fish in 2 lakes</td>
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<tr>
<td>Observations</td>
<td>194</td>
<td></td>
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Table 5. Linear mixed-effect model of temperature at capture (last annuli) of adults lake trout (response variable). Columns present the estimated differences (positive or negative) of factor response variables with the model intercept, standard error (SE), degrees of freedom (df) and $p$-values for the factor. $p$-values in bold indicate significant differences ($p < 0.05$). The random parts show the number of tested groups and experimental units ($N_{grp}$) and total observations.

<table>
<thead>
<tr>
<th>Coefficient</th>
<th>Response</th>
<th>Estimate</th>
<th>SE</th>
<th>df</th>
<th>$p$-value</th>
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<td>Adult temperature at capture</td>
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<tr>
<td>Intercept</td>
<td></td>
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<td>11.19</td>
<td>19</td>
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<tr>
<td>Total length</td>
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<td>0.007</td>
<td>19</td>
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<td>0.02</td>
<td>0.01</td>
<td>19</td>
<td>0.06</td>
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<tr>
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<tr>
<td>C:N</td>
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<td>1.63</td>
<td>19</td>
<td><strong>0.02</strong></td>
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<table>
<thead>
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<th>Random parts</th>
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<tbody>
<tr>
<td>$N_{grp}$</td>
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<tr>
<td>Observations</td>
</tr>
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</table>
FIGURES CAPTIONS

Figure 1. Ecological synopsis of studied lakes, maps are showing geographical position and morphology of both lakes. Fish communities of both lake are depicted as fish silhouette (credit: Phylopic website) and common names (see text for scientific names) under each map. Temperatures by depth profiles of each lake are provided with lake area (ha), maximum depth ($Z_{max}$) and Secchi depth.

Figure 2. Difference between the occupied habitat temperature at the end of the first growing season and at the time of hatching (otolith core) of local (blue) and hybrid (green) young-of-the-year from lakes Louisa (circles) and McFee (triangles). The black dashed line is a linear regression ($\Delta T_{end \ of \ season} = -0.87 \ast T_{hatching} + 9.45, p = 0.002, R^2_{adj} = 0.40$)

Figure 3. Estimated thermal habitat of juvenile lake trout for the first to the sixth growing season. Different letters indicate a significant difference in post-hoc pairwise comparisons

Figure 4. Mean and standard deviation (error bars) of lake trout lifelong thermal by age class (4 to 18 years) in each genetic origin groups in lakes Louisa (left panel) and McFee (right panel). Different colors in each graph represent a different genetic origin; local (green), hybrid (yellow) and stocked (red). Only positive or negative error bars are shown to optimize visualization.

Figure 5. Estimated temperature ($^\circ$C) at time capture (last annuli) of adult lake trout in relation to C:N ratio (top panel) and proportion of individual genotype related to source of stocking population ($Q_{stocked}$, bottom panel). Different colors in each graph represent a different genetic origin; local (green), hybrid (yellow) and stocked (red). In top panel, symbols are depicting lake of capture, either Louisa (circle) or McFee (square).
Figure 1.
Figure 2.
Figure 3.
Figure 4.
Figure 5.