Quantifying parasitic swimbladder nematodes and their effect on the body condition of Lake Whitefish in Great Slave Lake.

by

Caitlyn Friesen

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Abstract

Parasites are known to impact the health of their hosts due in part to depleting available energy. Although *Cystidicola farionis* are common parasitic swimbladder nematodes of Lake Whitefish, it is difficult to ascertain how infection affects the host's health. It is essential to understand parasite and host biology to assess the effects of parasitic infection on the host's body condition. Given that of the *Cystidicola* species, only *C. farionis* has been identified in Lake Whitefish, it is predicted that the nematodes will be identified as such. Reproduction is a major driver of an individual's health, and female Lake Whitefish invest more energy in reproductive events than males. The Fulton's (K) condition index was used to assess somatic body condition, where the inclusion of gonad weight could potentially mask a sex-specific effect of parasite infection. The gonadosomatic index (GSI) was used to determine if *C. farionis* infection has a sex-specific effect on Lake Whitefish's body condition. Swimbladder nematodes were identified based on egg morphology and genetic sequencing of the 28S and ITS-2 regions of the ribosomal DNA. For the first time in Great Slave Lake, *C. farionis* was identified based on morphological and genetic analysis, and prevalence was estimated at 54% in mature Lake Whitefish with a mean intensity of infection of 9.74 nematodes per infected host. T-tests indicated that body condition did not differ between Lake Whitefish infected with *C. farionis* and those uninfected. Although, linear regression models revealed that as the intensity of infection increased, K values of mature Lake Whitefish and GSI for current-year spawning females declined. There did not appear to be a sex-specific effect of infection, although future examination of current-year spawning males

and immature Lake Whitefish could provide further insight into the impact of *C. farionis* infection.

Acknowledgements

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Introduction

Parasites use hosts to complete their life cycle, directly and indirectly impacting the host's health by depleting available energy, reducing body condition, and, in extreme cases, inducing mortality (Robar *et al.* 2011; Harmon *et al.* 2015; Maceda-Veiga *et al.* 2016; Sánchez *et al.* 2018). The depletion in available energy can be due to hosts re-allocating energy towards responses to parasite infection from other necessary functions, like growth (Robar *et al.* 2011). In addition to particular life history traits being affected, parasitism may have more general effects, such as negatively impacting overall body condition due to the pathological effects of infection (Black 1984; Faisal *et al.* 2010; Lagrue and Poulin 2015; Maceda-Veiga *et al.* 2016). To assess the effects of parasites on their hosts, it is essential to first accurately identify them, which can be especially challenging with nematodes (Moravec 2007; Jones 2008).

Cystidicola spp. are common parasitic swimbladder nematodes of commercially important, physostomous fishes within North America, Europe, and Asia (Ko and Anderson 1969; Lankester and Smith 1980; Miscampbell *et al.* 2004; Faisal *et al.* 2010; Arai and Smith 2016; Dmitryjuk *et al.* 2022). There are three diagnostic tools capable of delimiting between the two species, *Cystidicola farionis* and *Cystidicola stigmatura*: host specificity, egg morphology, and genetic sequencing (Lankester and Smith 1980; Black 1983; Miscampbell *et al.* 2004; Faisal *et al.* 2010). Due to the considerable overlap between geographic range and host specificity for *Cystidicola* spp., egg morphology is a relatively accessible and reliable diagnostic characteristic for delimiting between *Cystidicola* spp. (Lankester and Smith 1980; Black 1983; Miscampbell *et al.* 2004; Faisal *et al.* 2010). However, mature eggs can only be found loose in the environment or within gravid female nematodes, making egg procurement difficult (Miscampbell *et al.* 2004; Faisal *et al.* 2010). In addition, identification based on egg morphology can only be verified based on morphological descriptions and host specificity, which has led to previous misidentifications, such as the misidentification of *C. farionis* as *C. stigmatura* in Great Slave Lake (Skinker 1931; Rawson 1951; Black 1983; Miscampbell *et al.* 2004). Subsequently, DNA sequencing has recently emerged as the best method to verify the morphological identification of *Cystidicola* spp., with previous studies using the 28S and ITS-2 regions of the ribosomal DNA (rDNA) (Miscampbell *et al.* 2004; Dmitryjuk *et al.* 2002).

Although host specificity alone cannot reliably delimit between *C. farionis* and *C. stigmatura*, only *C. farionis* has been reported from Lake Whitefish (*Coregonus clupeaformis*; Miscampbell *et al.* 2004; Dmitryjuk *et al.* 2022). In the Laurentian Great Lakes, the prevalence of *C. farionis* in Lake Whitefish varies from 2% to 50% between populations, likely due to varying levels of exposure to intermediate hosts through habitat use and local prey availability (Knudsen *et al.* 2004; Faisal *et al.* 2010). Lake Whitefish become infected by *C. farionis* by ingesting freshwater amphipods and mysids (i.e., opossum shrimp) infected with the third-stage larvae (Black and Lankester 1980; Lankester and Smith 1980; Faisal *et al.* 2010; Dmitryjuk *et al.* 2022; Fig. 1). Upon ingestion, the nematode travels from the esophagus through the pneumatic duct and into the swimbladder (Black and Lankester 1980; Dmitryjuk *et al.* 2022). Subsequently, with a diet consisting mainly of the intermediate hosts for *C. farionis*, fish hosts who eat more are predicted to have a higher chance of infection, increasing the potential harmful impacts associated with increased parasite load (Amundsen *et al.* 2003; Faisal *et al.* 2010;

Dmitryjuk *et al.* 2022). Although, some studies of parasites in fish have found that host body condition increased with infection (Faisal *et al.* 2010; Harmon *et al.* 2015; Lagrue and Poulin 2015; Maceda-Veiga *et al.* 2016). Within Great Slave Lake, the mere presence of parasites within the commercial fish has not been described since the 1950s, and the prevalence and effect of the parasites on the body condition of fish in Great Slave Lake have not been described (Rawson 1951; Fuller 1955).

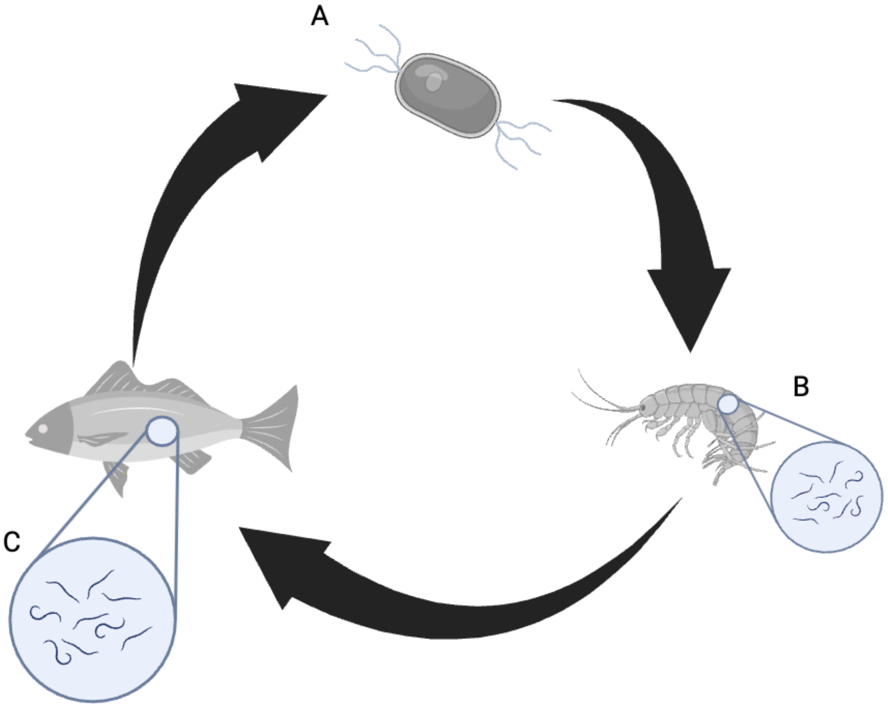

Figure 1: Simplified *Cystidicola farionis* life cycle showing transmission of a) *C. farionis* eggs to the intermediate host where they develop into the b) third-stage larvae, then to a definitive host where they develop into c) adults.

Reproductive strategy can also be a major driver of the temporal variability of an individual's overall health and, therefore, body condition (Jørgensen *et al.* 2006; Burness *et al.* 2008; Muir *et al.* 2014). As long-lived, iteroparous, benthic fish, Lake Whitefish exhibit a variety of strategies, such as intermittent spawning (Mikaelian *et al.* 2002; Carl and McGuiness 2006; Jørgensen *et al.* 2006; Wagner *et al.* 2010; Johnston *et al.* 2012; Morbey and Shuter 2013; Muir *et al.* 2014; Dixon *et al.* 2020). Rest periods are especially common in high latitude freshwater lakes, such as Great Slave Lake, due to the shorter growing seasons, limiting the energy allocated towards reproduction (Rideout *et al.* 2005; Morbey and Shuter 2013). This "skipped spawning" strategy is used when the success of future spawning events outweighs the loss of the current spawning event (Jørgensen *et al.* 2006; Johnston *et al.* 2012; Morbey and Shuter 2013; Muir *et al.* 2014; Dixon *et al.* 2020). Intermittent spawning is a strategy to account for poor conditions, given that current-year spawning individuals can reabsorb resources from gametes (resting) (Mikaelian *et al.* 2002; Rideout *et al.* 2005; Jørgensen *et al.* 2006; Morbey and Shuter 2013; Muir *et al.* 2014; Dixon *et al.* 2020). Therefore, in poor conditions, resting would be in the individual's best interest to ensure the success of future spawning events and reduced mortality (Rideout *et al.* 2005; Jørgensen *et al.* 2006; Dixon *et al.* 2020). Lake Whitefish may have evolved a quality control reproductive strategy to manage the unpredictable conditions of high-latitude freshwater lakes alongside intermittent spawning (Muir *et al.* 2014).

Lake Whitefish are proposed to follow the Reproductive Quality Control (RQC) hypothesis, making more frequent trade-offs from their somatic body condition or fecundity and more trade-offs of greater magnitude to ensure the quality of their offspring (Blukacz *et al.* 2010; Muir *et al.* 2014). If Lake Whitefish are parasitized, the RQC hypothesis supposes that these fish would allocate more energy towards the maintenance of gamete condition rather than overall body condition (Blukacz *et al.* 2010; Muir *et al.* 2014). Regardless of whether or not Lake Whitefish follow the RQC hypothesis, energy allocation towards spawning events differs between the sexes for Lake Whitefish (Blukacz *et al.* 2010; Dixon *et al.* 2020). Males invest less energy into reproduction and, as group spawners, are thought to only contribute towards reproduction with their sperm, competing through sperm competition (Burness *et al.* 2008; Dixon *et al.* 2020). Subsequently, it follows that parasitic infection could impact female Lake Whitefish more than males due to the difference in reproductive investment (Blukacz *et al.* 2008; Dixon *et al.* 2020).

This study aims to: (1) identify the species of swimbladder nematode utilizing morphological and genetic characteristics; (2) estimate the prevalence, mean abundance, and mean intensity of swimbladder nematodes in mature Lake Whitefish; (3) determine if there is a sex-specific effect of swimbladder parasites on the body condition of mature Lake Whitefish using two different indices, Fulton's condition index and the gonadosomatic index (GSI). I hypothesize that the swimbladder nematodes will be morphologically and genetically identified as *C. farionis*, given that of the two known *Cystidicola* species, only *C. farionis* has been identified from Lake Whitefish. I predict that host sex will not influence the effect of intensity on the body condition calculated with Fulton's condition index due to the inclusion of gonad weight, which would mask a sex-specific effect of infection. I also predict that female Lake Whitefish will have higher GSI values than males and that current-year spawners will have higher GSI values than resting individuals as the intensity of infection with swimbladder parasites increases.

Methods

Sample collection

Field sampling was performed in Hay River, NWT from six sites on Great Slave Lake in July of 2023, before the Lake Whitefish spawning season of October to December (Evans *et al.* 1987; Dixon *et al.* 2020). Mature Lake Whitefish were sampled and processed following the fisheries-independent gillnet study sampling protocol outlined Zhu *et al.* (2015). Round weight (± 1g), fork length (± 1mm), and gonad weight (± 1mm) were measured in the field. Sex was determined based on the presence or absence of eggs and the texture of the gonads. For example, mature females have large eggs and mature males have thick testes with lobing. Maturity was determined based on several factors, including size of eggs, thickness/juiciness of testes, lobing of testes, bruising, and size of gonads relative to body cavity. For instance, immature females have ovaries that are slightly scale-like in texture, whereas immature males have testes with a smooth texture. An individual is considered immature until their first spawning event. Current-year spawners will have minimal bruising and the gonads fill the body cavity. Any individual that has previously spawned but is not a current-year spawner is considered resting. Resting females have small eggs with moderate to heavy bruising and old/cloudy eggs may be present. Resting males have thin, lobed gonads with moderate to heavy bruising. The swimbladders were only sampled after maturity was determined, using the previously mentioned characteristics, and the swimbladder was observed as intact (i.e., without puncture wounds and inflated) to minimize the possibility of losing free nematodes into the body cavity.

Following our field sampling and assessment of maturity and sex, 100 swimbladders from mature Lake Whitefish were collected from 57 females and 43 males. Swimbladders were removed from the body of the fish whole and then cut open along their entire length. They were then placed in 70% ethanol and glycerine in double bagged Ziplock bags, after removing as many air bubbles as possible. They were stored flat at -20°C in the field and moved to -30°C in the lab. These precautions were taken to maximize the surface area of the swimbladder in contact with the ethanol for optimal parasite preservation.

A total of 10 nematode vouchers (i.e., nematode specimens collected for the analysis and documentation of morphological and genetic characteristics) were collected when there were visible nematodes in the swimbladder from 10 separate Lake Whitefish. Morphological and genetic analyses were conducted on the same nematode when eggs were present within the gravid females. The nematode voucher was preserved in 95% ethanol to improve the preservation of the DNA. All vouchers were stored at -20°C in the field and moved to -20°C in the lab.

Parasite identification

Nematodes were examined under light microscopy (400X magnification) and identified based on the diagnostic egg characteristics for *C. farionis* and *C. stigmatura* (Ko and Anderson 1969; Smith and Lankester 1979; Lankester and Smith 1980; Black 1983; Miscampbell *et al.* 2004; Faisal *et al.* 2010). Mature *C. farionis* eggs have polar filaments (Fig. 2a) and may also have lateral filaments or lobes (Fig. 2b) (Miscampbell *et* *al.* 2004; Faisal *et al.* 2010). In contrast, mature *C. stigmatura* eggs only have lateral lobes (Fig 2c) (Miscampbell *et al.* 2004; Faisal *et al.* 2010; Arai and Smith 2016).

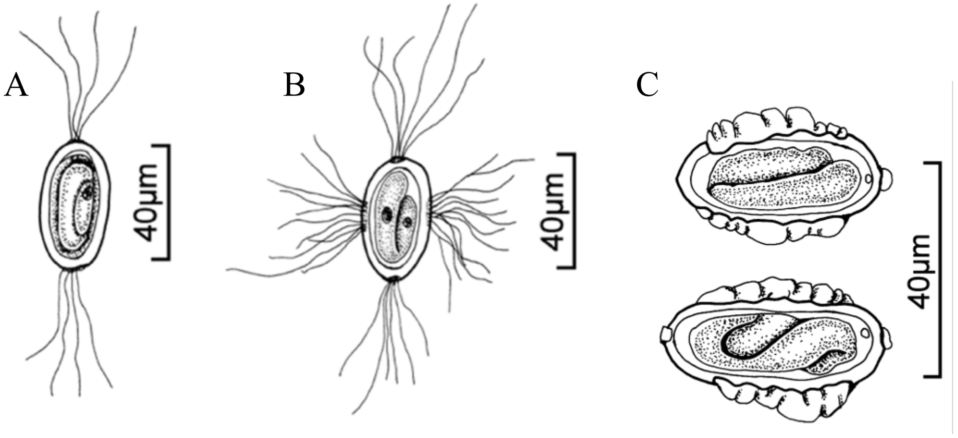

Figure 2: Fully developed eggs of *Cystidicola farionis* with a) only polar filaments and b) both lateral and polar filaments and *Cystidicola stigmatura* with c) lateral lobes. Modified from Arai and Smith (2016).

DNA was extracted from whole single nematodes torn into three pieces with the DNeasy Blood and Tissue Kit (QIAGEN, Hilden, Germany) with some modification to the X protocol. These included lysing the DNA overnight at 55°C at 500rpm using a Thermomixer (Eppendorf). Moreover, the optional step was performed to remove the RNAses and the final elution was performed twice with the same elute in an attempt to increase the concentration of DNA.

PCR amplification of the 28S region was performed in 25µl reactions containing 1X Colorless GoTaq Flexi Buffer, 2.0mM $MgCl_2$, 0.2mM dNTPs, 0.6µM of each primer (F-Nem28SF and R-Nem28SR; Nadler *et al.* 2000), 1.25U GoTaq DNA polymerase, 14.25µl water, and 1µl template DNA. PCR conditions consisted of one cycle of 95°C for 2 minutes (denaturation) followed by 30 cycles of 95°C for 30 seconds (denaturation), 55°C for 30 seconds (annealing), and 72°C for 1 minute (extension) and one cycle of 72°C for 7 minutes (extension).

PCR amplification of the ITS-2 region was performed in 25µl reactions containing 1X Colorless GoTaq Flexi Buffer, 2.5mM $MgCl_2$, 0.2mM dNTPs, 0.4µM of each primer (F-NC13 and R-NC2; Miscampbell *et al.* 2004), 1.25U of GoTaq DNA polymerase, 14.25µl water, and 1µl template DNA. PCR conditions consisted of one cycle of 95°C for 2 minutes (denaturation) followed by 35 cycles of 95°C for 50 seconds (denaturing), 55°C for 25 seconds (annealing), and 72°C for 1 minute (extension) and one cycle of 72°C for 7 minutes (extension).

Gel electrophoresis was performed in 2% agarose gel with 1X TAE buffer with 2.5µl aliquots of the PCR products to visualize the bands. PCR products were purified with the GeneJET PCR Purification Kit (ThermoFisher Scientific, Baltics, UAB) with some modification to the purification protocol. These modifications include using 20µl of the Binding Buffer and eluting with 25µl of water.

Sequencing was performed by the Hospital for Sick Children with an ABI 3730XL Sanger sequencer (ThermoFisher Scientific, Baltics, UAB). Alignments were done in MEGA11 (version 11.0.13). Sequences were then compared to DNA sequences in the GenBank database (https://www.ncbi.nlm.nih.gov/genbank/, accessed on 4 April, 2024) with the blastn algorithm (https://blast.ncbi.nlm.nih.gov/Blast.cgi, accessed on 4 April, 2024).

Data analysis

To isolate nematodes for counting, each swimbladder was strained in a 500µm sieve. Counts were conducted using a stereomicroscope set at 7-120X magnification. Prevalence of infection with swimbladder parasites was calculated by dividing the number of infected Lake Whitefish by the total number of sampled Lake Whitefish and multiplying by 100 for the percentage of the population infected with swimbladder nematodes (Bush *et al.* 1997). Mean abundance was calculated by dividing the total number of nematodes by the total number of Lake Whitefish, including the non-infected individuals (Bush *et al.* 1997). Intensity of infection is the number of nematodes within an individual (Bush *et al.* 1997). The mean intensity of infection was calculated by dividing the total number of nematodes by the number of infected Lake Whitefish (Bush *et al.* 1997). The nematode counts include any nematodes that may have been removed for morphological and genetic vouchers.

Body condition of the Lake Whitefish was determined using Fulton's condition index (K) which was calculated based on the round weight (W) and fork length (L) of the Lake Whitefish using the following formula (eq. 1).

$$(1)\ K = \frac{W}{L^3} \times 100{,}000$$

The K value can provide insight into the individual's overall condition and indirectly measure the factors that affect an individual's condition (Rennie and Verdon 2008; Zhu *et al.* 2015). K values of less than one indicates poor body condition and more than one indicates good body condition.

The gonadosomatic index (GSI) was also used to provide more insight into an individual's energy allocation (Lumb *et al.* 2007; Johnston *et al.* 2012; Dixon *et al.* 2020;

Betsy *et al.* 2021). GSI was calculated based on the gonad weight (GW) and round weight (W) of the Lake Whitefish using the following formula (eq. 2).

$$(2)\ GSI = \frac{GW}{W} \times 100$$

The GSI value indicates the proportion of the round weight composed of gonad mass and can be used to indicate sexual maturity, fecundity, and reproductive investment of the individual (Lumb *et al.* 2007; Dixon *et al.* 2020; Betsy *et al.* 2021). Only current-year spawners would follow the RQC hypothesis, whereas resting individuals compromise the condition of their gonads for their overall health and survival (Jørgensen *et al.* 2006). Therefore, GSI was analyzed separately for current-year spawners and resting individuals.

All statistical analyses were conducted in R (Version 2023.12.1+402 (2023.12.1+402)). Two-sample t-tests were used to determine if there was a significant difference between: 1) the mean K value based on infection status for mature Lake Whitefish, 2) the mean K value of the two sexes for mature, infected Lake Whitefish, 3) the mean GSI between current-year spawning and resting, infected, female Lake Whitefish, and 4) the mean GSI between uninfected and infected resting, female Lake Whitefish. Because the sample variations were unequal, a Welch's two-sample t-test was used to compare: 1) the mean GSI between uninfected and infected current-year spawning, female Lake Whitefish and 2) the mean GSI of the two sexes for infected, resting Lake Whitefish.

Linear regression models were used to compare the intensity of infection with body condition indices. The K and GSI values were log transformed to meet assumptions of normality and homoscedasticity (McDonald 2014). Linear regression models were used to compare: 1) intensity of infection with the log of K for mature, infected Lake

Whitefish, 2) the log of GSI for mature, infected, female Lake Whitefish, and 3) the log of GSI for resting, infected Lake Whitefish.

Results

The nematodes were identified as *C. farionis* due to the presence of polar and lateral filaments on the mature eggs (Fig. 3). All nine 28S sequences recovered were identical to one another and had 100% identity match to 28S sequences in *C. farionis*, with query cover ranging from 39% to 100%, including *C. farionis* from *Salmo trutta* in Switzerland (MT086834.1) and *Osmerus eperlanus* in Poland (OM691417.1). The nine ITS-2 sequences recovered were identical and only had two results from the GenBank database, with 100% query cover for both results. The ITS-2 sequences had a 98.37% identity match to *C. stigmatura* (AY161297.1) and a 100% identity match to *C. farionis* (AY161296.1).

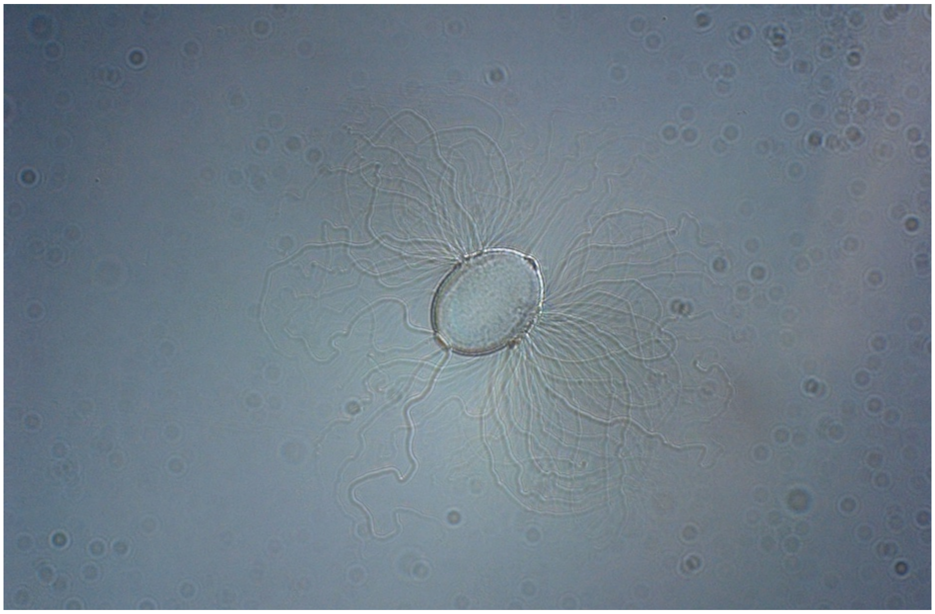

Figure 3: Mature *Cystidicola farionis* egg with polar and lateral filaments.

Nematodes were found in the swimbladders of 54% (54/100) of the swimbladders of Lake Whitefish collected from Great Slave Lake. Nearly 55% (29/53) of females and 53% (25/47) of males were infected with *C. farionis*. The number of nematodes collected within a single swimbladder ranged from 1 to 60 within a single swimbladder. Almost all nematodes were found free within the swimbladder cavity, except one nematode was found between the layers of the membranes. The mean abundance was 5.26 ($n = 100, SD = 10.5$) nematodes per Lake Whitefish, including the uninfected individuals. The mean intensity of infection was 9.74 ($n = 54, SD = 12.7$) nematodes per infected Lake Whitefish.

K values ranged from 0.99 to 1.88 for all Lake Whitefish sampled, with only one individual, a current-year spawning female, with a normal body condition and all others in good condition. Uninfected Lake Whitefish had K values ranging from 0.99 to 1.88 with a mean of 1.46 ($n = 46, SD = 0.154$) and K values for Lake Whitefish infected with *C. farionis* ranged from 1.23 to 1.70 with a mean of 1.44 ($n = 54, SD = 0.106$; Fig. 4). There were no significant differences in K between infected and uninfected Lake Whitefish ($t = -0.573, df = 98, P = 0.568$).

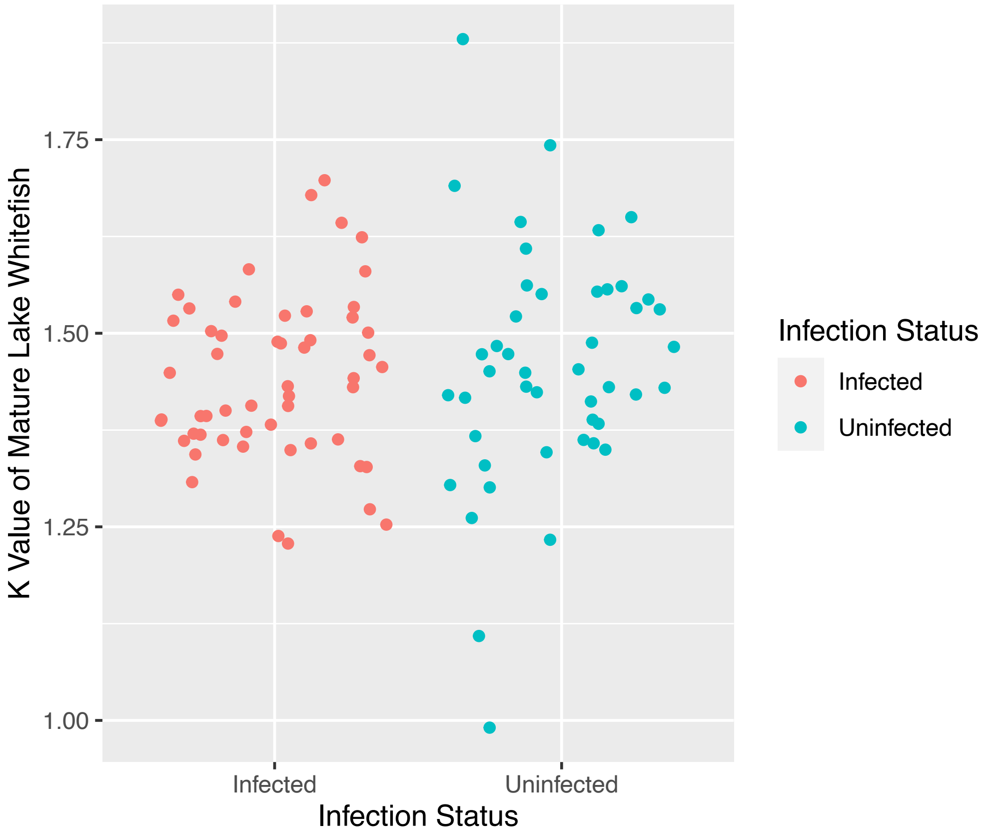

Figure 4: K values did not differ (P>0.05) between mature Lake Whitefish infected with *Cystidicola farionis* (n=54) and uninfected (n=46).

As intensity of infection increased, the log of K for mature, infected Lake Whitefish significantly decreased ($P = 0.0409$; Fig. 5), where the log of K declined by -7.04×10^{-4} ($SD = 0.000336$) for every additional nematode present. K values did not differ between mature, infected female and male Lake Whitefish ($t = 0.728, df = 52, P = 0.470$). Female Lake Whitefish infected with *C. farionis* had K values ranging from 1.24 to 1.70, with a mean of 1.45 ($n = 29, SD = 0.112$). Infected males had K values ranging from 1.23 to 1.68, with a mean of 1.43 ($n = 25, SD = 0.100$).

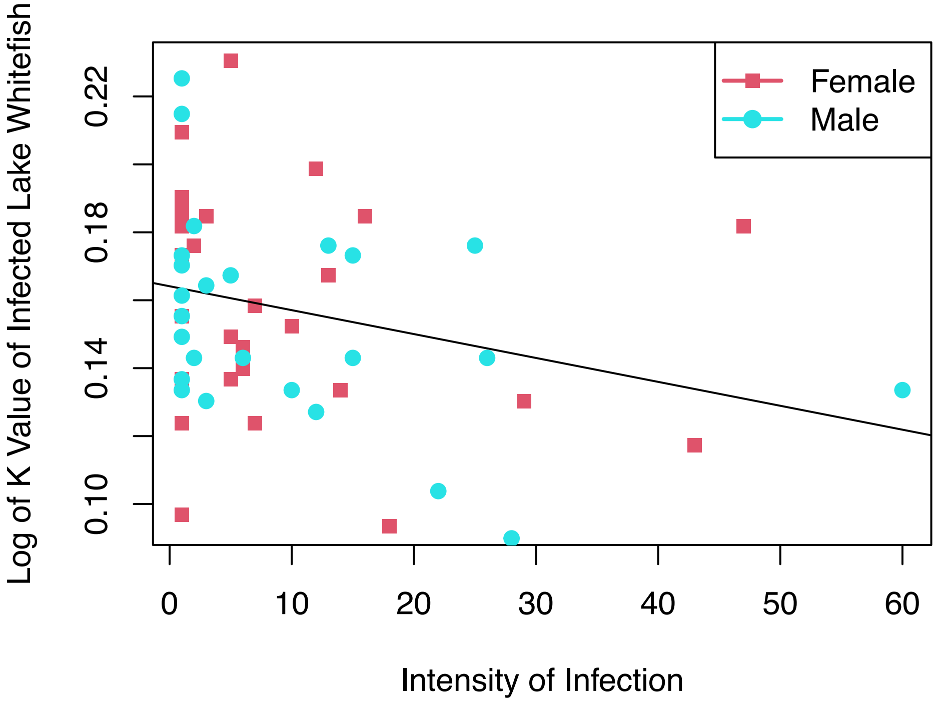

Figure 5: A negative relationship between intensity of infection with *Cystidicola farionis* and log K value for mature, infected Lake Whitefish (n=54).

Nearly 65% (37/57) of females and 19% (8/43) of males were current-year spawners. GSI values could not be effectively compared between current-year spawning female and male Lake Whitefish due to the low sample size of current-year spawning males. The mean GSI for current-year spawning, female Lake Whitefish ($n = 36$) was significantly higher than that for resting, female Lake Whitefish ($n = 10$; $t = 4.78, df = 26, P < 0.0001$; Fig. 6). Current-year spawning females had GSI values ranging from 1.45% to 5.90%, with a mean of 3.63% ($n = 36, SD = 0.973\%$). Resting females had GSI values ranging from 0.34% to 5.84%, with a mean of 1.89% ($n = 19, SD = 1.46\%$).

Figure 6: Current-year spawning, female Lake Whitefish (n=36) had higher GSI values (%) than the resting females (n=19).

GSI values did not differ between the current-year spawning, female Lake Whitefish that were infected with *C. farionis* ($n = 18$) and those that were uninfected ($n = 18$; $t = 1.02, df = 30.3, P = 0.315$). Infected, current-year spawning females had GSI values ranging from 1.45% to 5.90%, with a mean of 3.79% ($n = 18, SD = 1.23\%$). Uninfected, current-year spawning females had GSI values ranging from 1.98% to 5.05%, with a mean of 3.46% ($n = 18, SD = 0.782$). GSI values did not differ between the resting, female Lake Whitefish that were infected with *C. farionis* ($n = 10$) and those that were uninfected ($n = 9$; $t = -0.669, df = 17, P = 0.512$). Infected, resting females had

GSI values ranging from 0.34% to 3.70%, with a mean of 1.67% ($n = 10, SD = 1.11\%$). Uninfected, resting females had GSI values ranging from 0.51% to 5.84%, with a mean of 2.13% ($n = 9, SD = 1.80\%$). As intensity of infection increased, the log of GSI (%) for infected, current-year spawning, female Lake Whitefish significantly decreased ($P = 0.0270$; Fig. 7), where the log of GSI decreased by 0.00524% for every additional nematode. The log of GSI (%) did not differ as intensity of infection increased for infected, resting, female Lake Whitefish ($P = 0.724$).

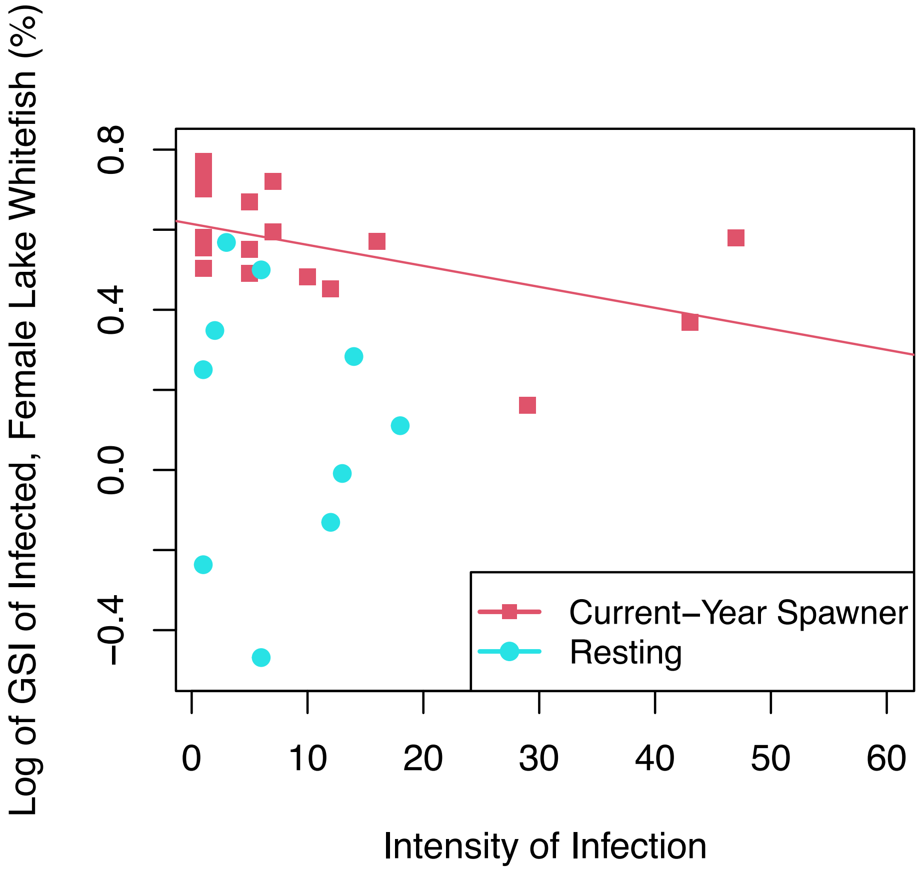

Figure 7: A negative relationship between intensity of infection with *Cystidicola farionis* and log GSI (%) for the current-year spawning, female Lake Whitefish (n=18), but log GSI (%) for resting, female Lake Whitefish (n=10) did not differ.

Of the resting Lake Whitefish, 11 females and 22 males were infected with *C. farionis*, allowing for the comparison of the effect of intensity of infection on GSI between resting, infected, female and male Lake Whitefish. Infected, resting, male Lake Whitefish had GSI values ranging from 0.45% to 1.80%, with a mean of 1.00% ($n = 17, SD = 0.350\%$). The mean GSI values (%) did not differ between infected, resting, female and male Lake Whitefish ($t = 1.84, df = 10.1, P = 0.0950$). The log of GSI (%) did not differ as intensity of infection increased for infected, resting Lake Whitefish ($P = 0.475$; Fig. 8).

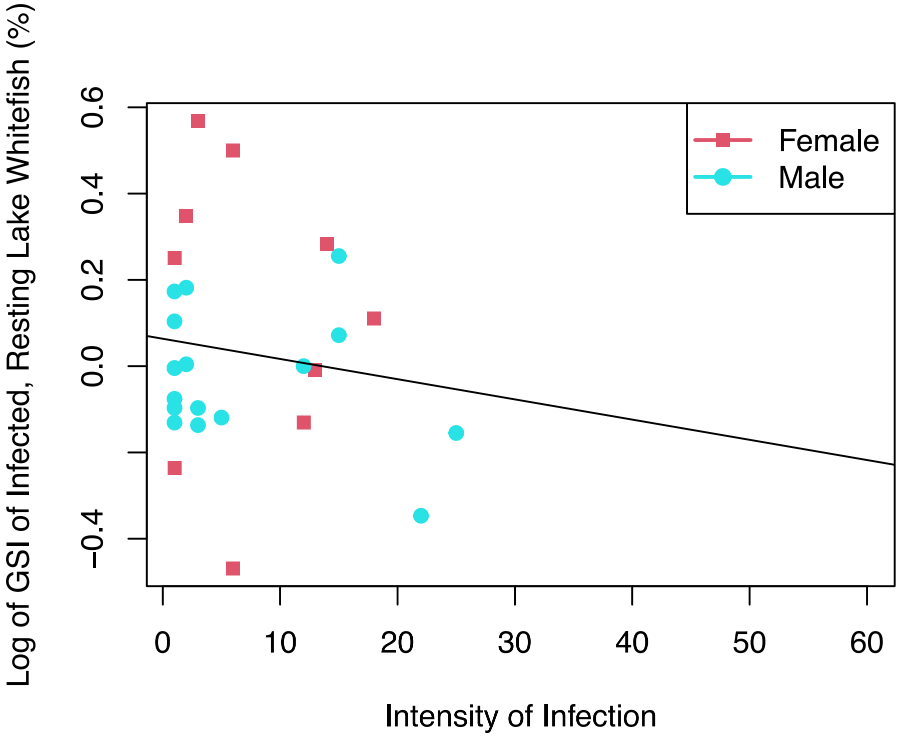

Figure 8: Log GSI (%) did not change as intensity of infection with *Cystidicola farionis* increased for resting, female (n=10) and male (n=17) Lake Whitefish.

Discussion

Morphologically, the eggs from Great Slave Lake are consistent with the *C. farionis* collected from Lake Whitefish from Ontario in that they have polar and lateral filaments (Lankester and Smith 1980; Faisal *et al.* 2010). However, they differ from *C. farionis* eggs from Lake Whitefish found in British Colombia, Finland, and Poland in that the latter eggs lack later filaments (Miscampbell *et al.* 2004; Dmitryjuk *et al.* 2022). Thus, egg morphology appears to vary geographically and could be explained by isolation from glacial refugia (Foote *et al.* 1992).

This study is the first to estimate the prevalence of *C. farionis* in Lake Whitefish within Great Slave Lake. Relative to other studies of *C. farionis* in Lake Whitefish, a prevalence of 54% is high, though a mean intensity of 9.74 nematodes per infected Lake Whitefish is low compared to 2.24% (Faisal *et al.* 2010) and 54.05% (Dmitryjuk *et al.* 2022). Typically, a high prevalence of infection with *C. farionis* corresponds to high intensities of infection in Lake Whitefish (Amundsen *et al.* 2003; Faisal *et al.* 2010; Dmitryjuk *et al.* 2022). The lower intensity of infection found in our study could be due to the reduction of infected intermediate hosts, as Lake Whitefish consume both the parasite and the prey (Kołodziej-Sobocińska 2019). Subsequently, this would result in the initial infection with *C. farionis* among many Lake Whitefish, reducing the availability of infected intermediate hosts and minimizing the number of Lake Whitefish with high parasite loads (Kołodziej-Sobocińska 2019).

Another explanation for the relatively low intensity of *C. farionis* is the specialized diet of the Lake Whitefish within Great Slave Lake (Knudsen *et al.* 2004). As the ice retreats, a pulse of resources becomes available, causing cascading effects throughout the ecosystem (Ji *et al.* 2013; Weber and Brown 2018). Pulsed resource events are increasingly common as latitude increases, resulting in an increased abundance of primary consumers like amphipods and mysids (Ji *et al.* 2013; Weber and Brown 2018). As Lake Whitefish preferentially feed on amphipods when available, the Lake Whitefish of Great Slave Lake may have a more specialized diet than those at lower latitudes, such as within the Great Lakes (Pothoven *et al.* 2001; Pothoven *et al.* 2006; Lumb *et al.* 2007). As Lake Whitefish with more generalized diets accumulate higher aggregations of parasites, a more specialized diet would decrease infection intensity (Knudsen *et al.* 2004).

The negative relationship between the intensity of infection and overall body condition is likely due to the cumulative negative impact of increased parasite load (Robar *et al.* 2011; Maceda-Veiga *et al.* 2016). *C. farionis* feeds on Lake Whitefish blood and tissue, depleting available resources (Dmitryjuk *et al.* 2022). In addition, increased parasite load could increase the cost of mounting an immune response, resulting in less energy for somatic growth (Robar *et al.* 2011; Maceda-Veiga *et al.* 2016).

The intensity of infection with *C. farionis* was not found to have a sex-specific effect on K value. Infection is directly related to prey availability and consumption of infected intermediate hosts, regardless of sex (Knudsen *et al.* 2004). Subsequently, if infection with *C. farionis* and environmental conditions are consistent for both sexes, then energy allocation for female and male Lake Whitefish may only differ in reproduction (Burness *et al.* 2008; Blukacz *et al.* 2010; Dixon *et al.* 2020). Fulton’s condition index provides insight into an individual’s somatic condition but cannot infer reproductive investment, potentially missing any sex-specific effects of infection (Rennie and Verdon 2008; Zhu *et al.* 2015).

Although the increased intensity of infection with *C. farionis* reduced the body condition of infected Lake Whitefish, the mean K value of infected Lake Whitefish did not differ from that of uninfected Lake Whitefish. This result suggests that Lake Whitefish increased their consumption of amphipods and mysids due to a pulsed resource event, which improved their body condition (Pothoven *et al.* 2001; Knudsen *et al.* 2004; Pothoven *et al.* 2006; Ji *et al.* 2013; Weber and Brown 2018). Subsequently, any negative impacts from infection would be masked by the energy gained from the resource pulse (Pothoven *et al.* 2001; Knudsen *et al.* 2004; Pothoven *et al.* 2006; Ji *et al.* 2013; Maceda-Veiga *et al.* 2016; Weber and Brown 2018).

The GSI of current-year spawning female Lake Whitefish was nearly double that of resting females, representing the increased reproductive investment of current-year spawning females (Lumb *et al.* 2007; Dixon *et al.* 2020; Betsy *et al.* 2021). GSI did not differ between uninfected and infected current year spawning female Lake Whitefish. Although the intensity of infection with *C. farionis* increased, the GSI of current-year spawning female Lake Whitefish decreased, likely due to heavily infected Lake Whitefish undergoing atresia to reabsorb oocytes in response to the harmful impacts of increasing parasite load (Tyler and Sumpter 1996; Mikaelian *et al.* 2002; Rideout *et al.* 2005; Jørgensen *et al.* 2006; Sànchez *et al.* 2018). The decreased GSI of current-year spawning females could support the RQC hypothesis, where a reduced GSI would reflect a reduction in fecundity to maintain gamete quality (Muir *et al.* 2014).

GSI for resting Lake Whitefish did not differ regardless of infection status, intensity of infection or sex. During rest periods, Lake Whitefish do not allocate energy towards reproduction, regardless of sex (Jørgensen *et al.* 2006; Johnston *et al.* 2012; Morbey and Shuter 2013; Muir *et al.* 2014; Dixon *et al.* 2020). Subsequently, GSI for resting individuals is already at a minimum and cannot worsen due to worsening conditions.

Conclusion

For the first time, *C. farionis* was identified and estimated to have a prevalence of 54% within mature Lake Whitefish in Great Slave Lake with morphological and genetic analyses. This study found evidence of a negative relationship between intensity of infection and host body condition for both the Fulton's condition index and the GSI. Although there did not appear to be a sex-specific effect, further examination of current-year spawning male and immature Lake Whitefish should be pursued.

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