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EVIDENCE OF A PUTATIVE NOVEL SPECIES OF AVIAN SCHISTOSOME INFECTING *PLANORBELLA TRIVOLVIS*

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KEY WORDS

Digenean
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Helisoma trivolvis
Swimmer's itch
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ABSTRACT

Freshwater gastropods of the genera *Lymnaea* Lamarck, 1799, *Physa* Draparnaud, 1801, *Gyraulus* Charpentier, 1837, *Radix* Montfort, 1810, and *Stagnicola* Jeffreys, 1830 are considered suitable intermediate hosts for avian schistosomes. A large trematode biodiversity survey performed across 3 yr on 6 lakes in Alberta confirmed 3 already-reported snail hosts for 7 North American avian schistosomes; however, the cytochrome *c* oxidase subunit 1 (*COI*) nucleotide sequence from 1 cercarial sample (from a single specimen of *Planorbella trivolvis*) was distinct from all other *COI* schistosome sequences. As part of a simultaneous, comparable study of *P. trivolvis* by us in Michigan, we collected another cercarial type from 6 lakes that was 99% similar (*COI*) to the aforementioned cercarial type. Phylogenetic analyses of the *COI* and *28S rDNA* genes recovered the former cercaria in a clade of avian schistosomes. In Michigan, the feces of a Canada goose (*Branta canadensis* Linnaeus, 1758) had a miracidium with an identical *COI* nucleotide sequence. Preliminary swimmer's itch and cercarial emergence studies were performed to determine if the cercariae could cause swimmer's itch and to study the emergence pattern as compared with species of *Trichobilharzia* Skrjabin and Zakharow, 1920.

Members of the Schistosomatidae Weinland, 1858 infect avian or mammalian hosts as the definitive hosts in their life cycles (Cort, 1950; Blair and Islam, 1983; Brant and Loker, 2009a, 2009b). Schistosome eggs are released from the definitive host and hatch into miracidia, which go on to penetrate an aquatic snail. Within the snail, the miracidia develop into sporocysts, which asexually produce the free-living larval stage, cercariae. The cercariae emerge from the snail (Cort, 1928) and must go on to find a suitable avian or mammalian definitive host to continue the life cycle.

When schistosomes penetrate humans, they can cause a variety of complications. Species of *Schistosoma* Weinland, 1858 cause the disease schistosomiasis, which is endemic to tropical countries in Africa, Asia, and South America (World Health Organization Department of Communicable Disease Prevention, Control and Eradication, 1984). However, globally, most avian schistosomes and some cattle mammalian schistosomes (Horák et al., 2015) are responsible for cercarial dermatitis, known colloquially as swimmer's itch, with most of the research having been undertaken in North America and Europe (Cort, 1950; Blankespoor and Reimink, 1991; Horák et al., 2015; Gordy et al., 2018; Rudko et al., 2018, 2019). Snails of *Lymnaea, Physa, Gyraulus, Radix,* and *Stagnicola* have been considered the major drivers of swimmer's itch in the Northern Hemisphere and are ubiquitous (Horák et al., 2015). Given the efforts to understand the epidemiology of swimmer's itch, it is remarkable that over the century of screening a diversity of snails in North America for schistosomes, there has been no report of a schistosome infecting *Planorbella trivolvis* (=*Helisoma trivolvis*) Say, 1817, which is a relatively common snail to encounter. This is noteworthy considering that species of *Planorbella* and *Biomphalaria* Preston, 1910, the snail responsible for transmitting most cases of human schistosomiasis, are sister taxa (Morgan et al., 2002). According to the Natural History Museum's Host–Parasite database (Gibson et al., 2005; http://www.nhm.ac.uk/research-curation/scientific-resources/taxonomy-systematics/host-parasites/), there are no reports of species of *Planorbella* or *Helisoma* hosting undescribed or even described species of avian or mammalian schistosomes (e.g.,

Trichobilharzia spp., *Dendritobilharzia* spp. Skrjabin, 1920, *Gigantobilharzia* spp. Odhner, 1910, *Anserobilharzia* sp. Farr and Blankemeyer, 1956, or *Schistosomatium* sp. Tanabe, 1923) that are known to cause cercarial dermatitis (Cort, 1950; Hoeffler, 1974; Kolářová et al., 1999; Brant and Loker, 2009b; Brant et al., 2013; Soldánová et al., 2013).

Two parallel studies in North America, 1 in Michigan ongoing since June 2016 (Rudko et al., 2018, 2019) and 1 that was completed in Alberta (Gordy et al., 2016; Gordy and Hanington, 2019), found schistosome-like cercariae emerging from *P. trivolvis* snails. The cercariae resembled those of schistosomes, exhibiting eyespots and bifurcated tails (Cort, 1950). Since their morphological features alone could not be used to identify the cercariae, it was necessary to perform phylogenetic analyses that would determine if this was a known schistosome species discovered in a novel snail host. For instance, another planorbid genus, *Gyraulus*, is host to a diversity of avian schistosomes (Laman et al., 1984; Brant and Loker, 2009a, 2009b; Aldhoun et al., 2012; Brant et al., 2013), so one of those species might have infected a *P. trivolvis* snail. The other, more interesting possibility was that this parasite was a novel species in a genus of snails thus far never reported to host schistosomes.

It is often difficult to identify digeneans for several reasons, including life cycle gaps that make it challenging to link cercariae to adult worms, trouble obtaining adult worms from definitive host species, and similar morphological features that complicate identification (e.g., Kolářová et al., 1999; Brant and Loker, 2009b; Gordy et al., 2016). Acknowledging these challenges, this study aimed to morphologically and genetically characterize the putative novel schistosome found in *P. trivolvis* and to identify the definitive host.

MATERIALS AND METHODS

Collections

Snail collections in Michigan were performed as described by Rudko et al. (2019), and collection protocols for the trematode biodiversity study performed in Alberta are outlined by Gordy et al. (2016). For the ongoing trematode survey in Michigan, snail species collected included *Stagnicola emarginata*, *Lymnaea* sp., *Physa integra*, *Pleurocera* sp. Rafinesque, 1818, *Planorbella trivolvis*, *Marstonia* sp. Baker, 1926, *Campeloma* sp. Rafinesque, 1819, *Viviparus* sp. Montfort, 1810, and *Gyraulus* sp. The snails collected in Michigan were identified with the help of the key from Burch (1982) by an experienced researcher who has studied swimmer's itch-causing parasites and their hosts in the area (Blankespoor and Reimink, 1991; Verbrugge et al., 2004; Froelich et al., 2019; Rudko et al., 2018, 2019). The 2019 Michigan collection season was the first year that snail collections started in early June, rather than late June as in previous years.

Cercariae of the putative novel schistosome were obtained from *P. trivolvis* snails collected from Walloon Lake, South Lake Leelanau, North Lake Leelanau, Glen Lake, Crooked Lake, and Long Lake in Michigan, USA and from Wabamun Lake in Alberta, Canada (Gordy and Hanington, 2019). DNA extraction, amplification, and sequencing were accomplished using the cercariae that emerged from the snails in Alberta and Michigan following established protocols (Gordy et al., 2016; Rudko et al., 2019). The mitochondrial cytochrome *c* oxidase subunit 1 (*COI*) sequence from Alberta was labeled avian schistosomatid sp. C (GenBank accession MH168793).

Fecal samples were collected from aquatic birds including mallards (*Anas platyrhynchos* Linnaeus, 1758), Canada geese (*Branta canadensis*), kingfishers (*Megaceryle alcyon* Linnaeus, 1758), common grackles (*Quiscalus quiscula* Linnaeus, 1758), common mergansers (*Mergus merganser* Linnaeus, 1758), hooded mergansers (*Lophodytes cucullatus* Linnaeus, 1758), and red-breasted mergansers (*Mergus serrator* Linnaeus, 1758). Samples were collected at Lake Charlevoix, South Lake Leelanau, Platte Lake, Long Lake, Crooked Lake, Torch Lake, Elk Lake, Lime Lake, and Glen Lake, all located in northwest Michigan. Fecal sample collection and miracidia hatching procedures were performed as outlined in Rudko et al. (2018).

Measurements were taken of the body length and width, tail stem length and width, furcae length, and anterior organ length and width of the putative schistosome cercariae from microscope images. Images were taken of a wet-mounted cercaria using a Zeiss Axio Scope.A1 (Zeiss, Toronto, Canada) and measurements were taken using ZEN (Zeiss) software (https://www.zeiss.com/microscopy/int/products/microscope-software/zen.html).

Preliminary swimmer's itch and cercarial emergence studies

To ascertain whether these putative novel schistosome cercariae from *P. trivolvis* could be etiologic agents of swimmer's itch, preliminary studies were performed to determine if the cercariae could cause swimmer's itch and to understand their pattern of emergence from the snail. For the swimmer's itch trials, water containing these cercariae was placed on the cleaned forearm of 4 researchers who had developed swimmer's itch in the past (University of Alberta Human Research Ethics Board Approval # Pro00055721). This detail is relevant because, for an unknown reason, not everyone who is exposed to swimmer's itch-causing cercariae develops the rash (Cort, 1928) and people become sensitized with repeated exposure to the cercariae (Olivier, 1949). A circle was drawn on the forearm with a permanent marker, and several live and active cercariae were pipetted from the water in which the snail had shed the cercariae. A water droplet was placed on the arm within the circle and the arm was held horizontal so that the water would not run off. After 20 min, the exposed area was washed and dried to remove cercariae and monitored for 72 hr.

A preliminary cercarial emergence study was performed. To do this, 11 previously screened and confirmed infected *P. trivolvis* collected from South Lake Leelanau, Michigan were placed individually into clear, lidded, 60-ml plastic cups with conditioned well water at 2200 hr. The water was collected in a bucket and left to sit overnight to become room temperature. The snails were then moved into a new cup at 2400 hr, 0530 hr, and then every 2 hr, with the last transfer occurring at 2200 hr. Crystal violet was added to each cup (3 µl/ml) after the snail was removed, and cercariae were counted at 24 hr after staining.

Sequencing

The mitochondrial *COI* and nuclear *28S rDNA* sequences from the putative novel schistosome cercariae that emerged from *P. trivolvis* collected in Michigan were analyzed to determine their phylogenetic relationship within the Schistosomatidae. DNA was

extracted and amplified using the *Trichobilharzia*-specific *COI* primers CO1F15 and CO1R15 (Brant and Loker, 2009a) following previously published protocols (Gordy et al., 2016) to obtain the *COI* sequences. For *28S* sequencing, U178 and L1642 primers (Lockyer et al., 2003) were used for amplification during polymerase chain reaction (PCR), with DIG12 and ECD2 (Tkach et al., 2003) added for sequencing. Before sequencing, a Truin Science PCR cleanup kit (catalog #: KTS1100) was used, following manufacturer protocols. A partial *28S* sequence for the putative novel schistosome was uploaded to GenBank (accession MT305665). To be certain of the identity of the snail that these parasites were utilizing as an intermediate host, a sample of snail DNA collected in July 2015 during the survey performed in Alberta (Gordy et al., 2016) was extracted, amplified, and Sanger sequenced (Macrogen Inc, South Korea) using the universal invertebrate primers LCO1490, and HCO2198 to obtain a *COI* sequence (Folmer et al., 1994). Following the same protocol that was used on the cercariae samples, DNA was also extracted from the schistosome miracidia that emerged from collected fecal samples. The amplification of the *COI* mitochondrial gene for these samples was accomplished using the same primers as those used for the cercariae, but because there were fewer miracidia per sample, the samples underwent 45 cycles of amplification. The sequences obtained from the cercariae and snail were input into Geneious version 11.0.6 (Kearse et al., 2012; http://www.geneious.com), trimmed, aligned, and the consensus sequences were queried against the BLASTn database (Zhang et al., 2000). The sequences with the highest percent nucleotide identity were selected, downloaded, and used to generate alignments and percent identity matrices to determine the identity of the sample organism.

Phylogenetic analyses

The *COI* and *28S* sequences were aligned separately and trimmed. From these alignments, Bayesian inference phylogenetic trees were created in Geneious using MrBayes (Huelsenbeck and Ronquist, 2001) with the generalized time reversible + gamma evolutionary model. Evolutionary models for both trees were chosen after exporting the trimmed nucleotide alignments into the program MEGA X (Kumar et al., 2018) and using the default parameters to determine the best DNA/protein model for the data sets. The outgroup used for the *28S* tree were *Ornithobilharzia canaliculata*, *Austrobilharzia variglandis*, and *Austrobilharzia terrigalensis* (Snyder, 2004; Brant et al., 2006) and from the *28S* tree results we selected the outgroup for the *COI* tree as 2 Schistosomatidae sp. sequences (Pinto et al., 2017). The parameters for both trees included 4 chains run at a chain length of 500,000, with the first 5,000 trees discarded as burn-in.

RESULTS

Collections

A total of 134 specimens of *P. trivolvis* were infected with the putative novel schistosome. Nine of 1,030 (0.87%) were infected during 2018, with 7 of these snails collected in June and the remaining 2 collected in July; 125 of 2,325 (5.38%) were infected during 2019, with 87 of the infected snails collected in July and 38 in June. To date, this parasite has not been found to emerge from these snails during August in Michigan or Alberta. In 2019, 359 fecal samples were collected from aquatic birds, of which 83 hatched schistosome miracidia from samples collected during July–September. A subset of the cercariae and DNA extraction were vouchered at the Museum of Southwestern Biology Division of Parasites MSB: Para:30827.

Cercarial measurements

Cercarial measurements from the present study are reported in Table I (n = 2 for all measurements except for those of the anterior organ where n = 1). Included for comparison are described and undescribed avian schistosomes from planorbid snails. Of the schistosome species used to create the phylogenetic trees (Figs. 1, 2), 15 of 37, including the putative novel schistosome, parasitize a planorbid snail as an intermediate host.

Swimmer's itch and cercarial emergence studies

Within 2 hr after the participants were exposed to the putative novel schistosome, a cercarial dermatitislike rash appeared in the form of a red papule on one of the participants, confirming that this parasite can cause swimmer's itch. As for the pattern of emergence for this trematode, it was found that the cercariae emerge sporadically during the daytime, but do not emerge when it is dark (Suppl. Fig. S1). Previous studies have indicated that the cercariae of some species of *Trichobilharzia* emerge during daylight hours and not after dark (Anderson et al., 1976; Soldánová et al., 2016).

Sequencing and phylogenetic analyses

The *COI* DNA sequences were obtained from 24 of the 83 fecal samples, with 1 of these samples matching avian schistosomatid sp. C from Gordy et al. (2018). The particular fecal sample that matched the cercariae sequences was collected in July at Lake Charlevoix, Michigan from a Canada goose (*B. canadensis*). This miracidia sample had a 99% nucleotide similarity to the *COI* sequences that originated from the cercarial samples collected from *P. trivolvis*.

Analysis of the parasitized snail tissue sample matched nucleotide sequences labeled in GenBank as *P. trivolvis* at 99.7%. In Alberta, this putative schistosome emerged from a single specimen of *P. trivolvis* from a total of 79 trematode-infected specimens collected during 3 collection seasons. In Michigan, the 2018 season was the first year that *P. trivolvis* was collected along with the regularly collected snail species known to contribute to swimmer's itch in Michigan.

The novel schistosome formed a clade with the avian schistosomes (Figs. 1, 2). The *COI* sequences from Michigan share a 99% nucleotide identity with the single sequence of the Alberta-collected specimen. This high nucleotide percent similarity has been interpreted in the past to indicate the conspecificity of specimens (Gordy et al., 2018).

DISCUSSION

After a century of screening snails for schistosomes, both for the devastating disease schistosomiasis and for swimmer's itch, herein is the first report of an avian schistosome from a *P. trivolvis* snail in North America. The phylogenetic results confirm that the putative schistosome from *P. trivolvis* is an avian schistosome that is

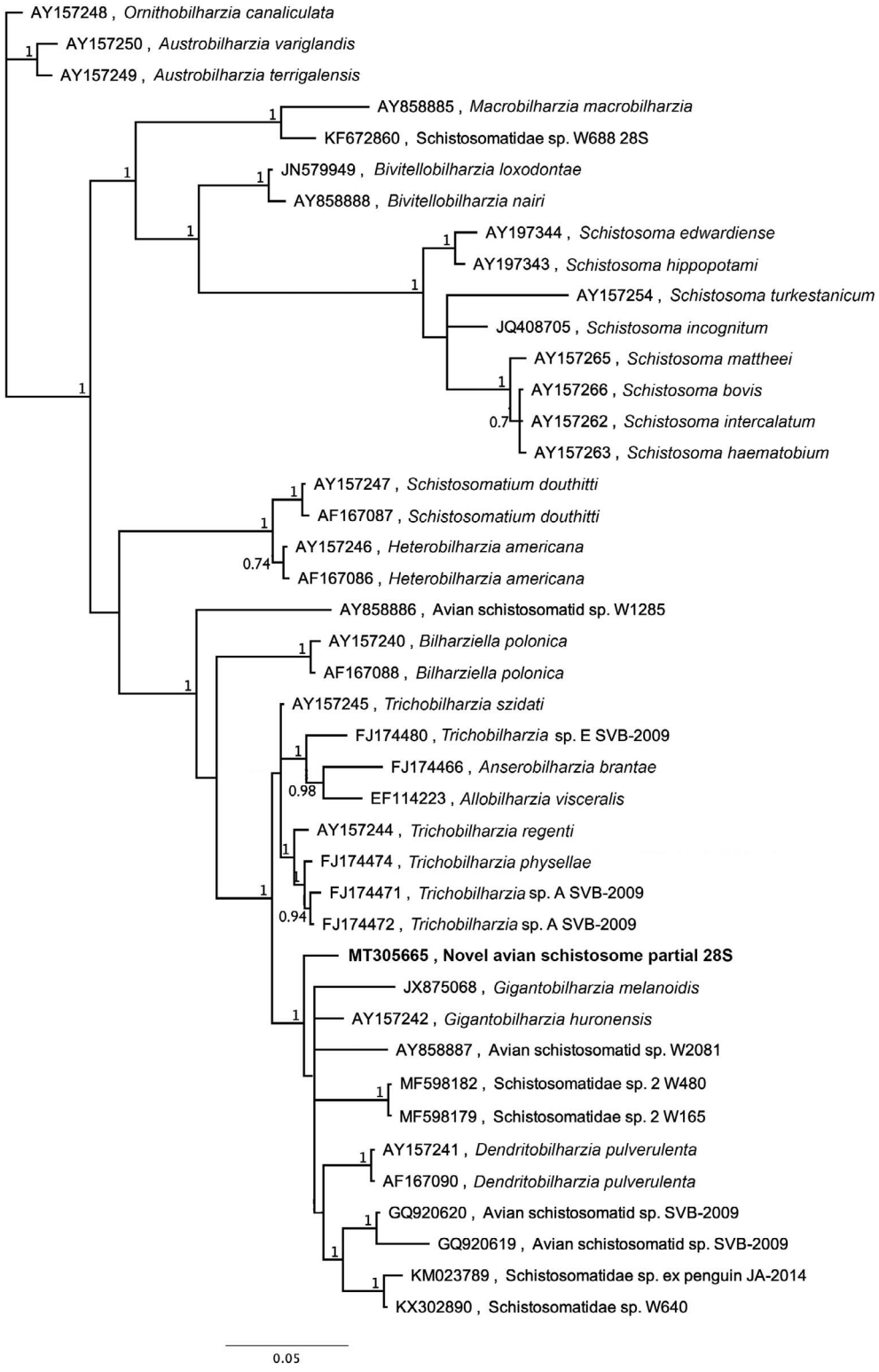

Figure 1. The *28S* phylogenetic tree of Schistosomatidae. Nodal support indicated by Bayesian posterior probabilities. GenBank accession numbers precede taxon names. The representative sample from this study is in bold.

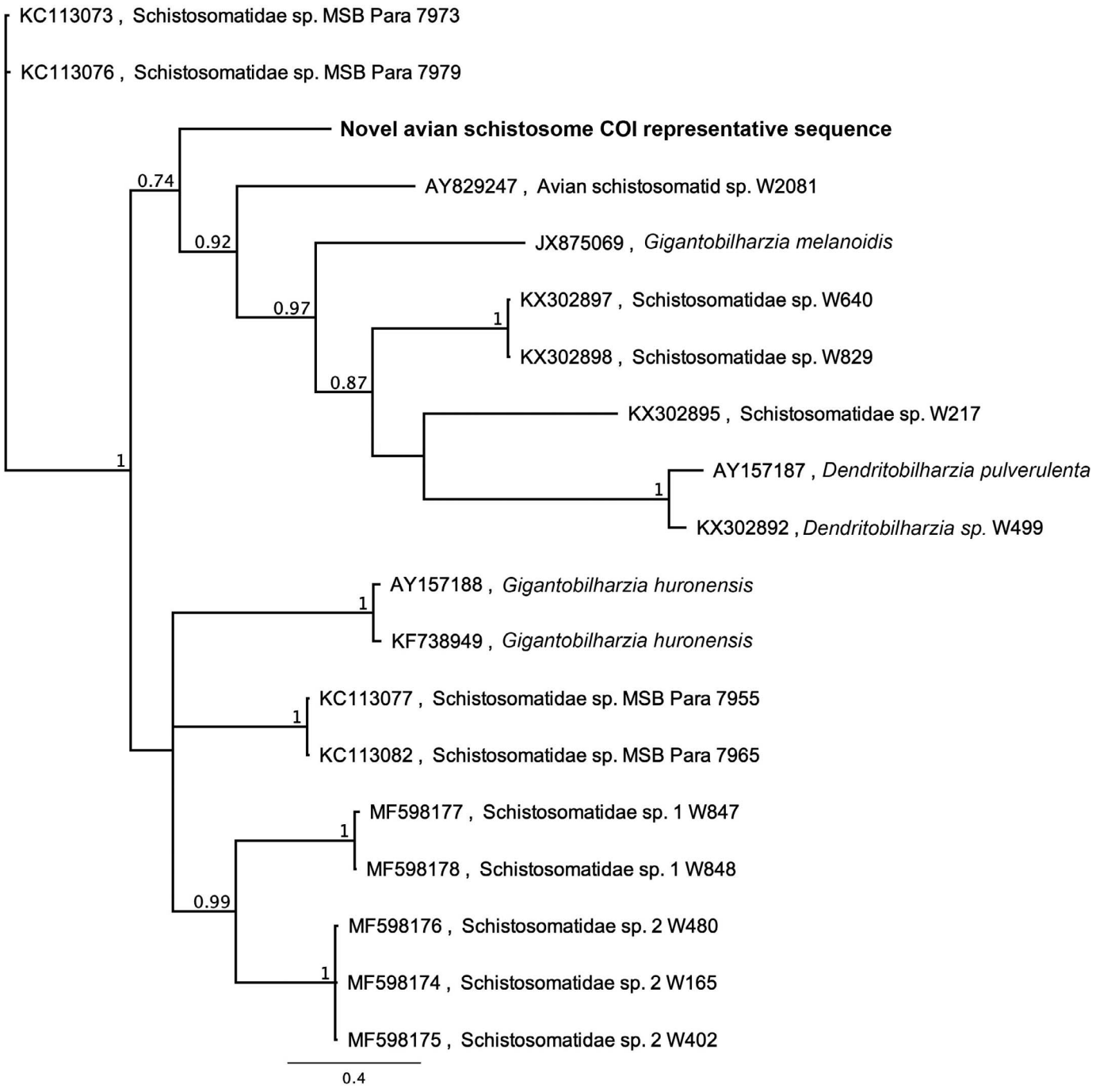

Figure 2. The cytochrome c oxidase 1 (*COI*) phylogenetic tree including members of the avian Schistosomatidae. Nodal support indicated by Bayesian posterior probabilities. GenBank accession numbers precede taxon names. The representative sample from this study is in bold.

conspecific between Alberta, Canada and Michigan, USA. Additionally, the miracidia hatched from feces of *B. canadensis* are conspecific with these cercariae. Therefore, the life cycle of this schistosome includes at least *P. trivolvis* as an intermediate host and a definitive host of a Canada goose, and possibly other commonly observed birds at these sites such as mallards and common mergansers. The cercariae emerge during daylight hours only and were confirmed to cause swimmer's itch, and thus likely contribute to the epidemiology of swimmer's itch where it is found.

There are other lineages of avian schistosomes that use planorbid snails, such as *Bilharziella polonica*, which is the most basal lineage of the large avian schistosome clade based on available taxa (Fig. 1). The diversity of planorbids used by avian schistosomes worldwide is remarkable: species in the genera *Gyraulus*, *Anisus* Studer, 1820, *Ceratophallus* Brown and Mandahl-Barth, 1973, *Segmentorbis* Mandahl-Barth, 1954, *Planorbis* Müller, 1773, *Biomphalaria*, *Indoplanorbis* Annandale and Prashad, 1921, and with this work, *Planorbella* (see Horák et al., 2015

Table I. Measurements for the putative novel avian schistosome plus other avian schistosome taxa from planorbid snails to compare across taxa from closely related snail hosts. Measurements are in micrometers. Abbreviations: L = length, W = width, M = cercariae measurement reference, SH = snail host reference(s). Values in parentheses represent the minimum and maximum measurements. "?" = sample size not indicated in referenced study. The information related to the putative novel schistosome is in bold.

			Cercariae measurements		
Schistosome	Locality	n	Body (L)	Body (W)	Tail stem (L)
Novel avian schistosome	**North America**	**2**	**(276–280)**	**(105–109)**	**(313–318)**
Schistosomatidae gen. sp. 2 (W403, W426)	North America	3	288 (267–300)	79 (73–88)	362 (346–398)
Schistosomatidae gen. sp. 1 (W847, W848)	South America	27	218 ± 33 (171–280)	57 ± 9 (41–82)	291 ± 20 (253–341)
Gigantobilharzia vittensis	Europe	17	286 ± 10 (265–298)	68 ± 5 (60–82)	383 ± 16 (358–415)
Avian schistosomatid sp. I6	Europe	13	275 ± 11	70 ± 4	345 ± 14
Gigantobilharzia gyrauli	North America	?	215 ± 3.3	73 ± 1.9	298 ± 2.8
Avian schistosomatid sp. W405	North America	1	125	65	300
Bilharziella polonica	Europe	?	(270–360)	(90–120)	(225–315)
Avian schistosomatid sp. W1285	Africa	4	(254–305)	(79–111)	(218–245)
Avian schistosomatid sp. W2081	Africa	1	286.3	85.6	446.9

for review). The measurements of the putative novel schistosome cercariae are closest in dimensions to Schistosomatidae sp. 1 (body length, tail length, and anterior organ length) and Schistosomatidae sp. 2 (body length), *Gigantobilharzia vittensis* (body length and anterior organ length), and avian schistosomatid sp. I6 (body length and anterior organ length) (Table I). These taxa are also the lineages with which the putative novel schistosome groups, but with no statistical support. Avian schistosomatid sp. I6 does not have an available *COI* sequence, but it grouped with Schistosomatidae sp. 1 and 2 from Pinto et al. (2017).

The schistosome from *P. trivolvis* and the Canada goose, though monophyletic, did not group with any other lineage of schistosome and, at this time, the group of schistosomes that is its closest relative is unknown. In the tree, it does cluster around species that are transmitted by smaller planorbid snails, such as species of *Gyraulus* and *Ceratophallus*, as well as samples from *Biomphalaria* collected from Brazil (Figs. 1, 2 and Table I). Without deeper node support for the clades, it is not possible to determine the diversification of schistosomes in planorbids relative to the other families of snails.

The putative novel schistosome appears to have a pan-North American distribution. Snails have been found infected with this schistosome in both Alberta and Michigan. This is likely because of the ubiquity of both the definitive host and the snail host. The spatial distribution of *B. canadensis* covers all of North America (Lincoln, 1935; Buhnerkempe et al., 2016), within which are defined 4 major routes for migratory birds: the Atlantic Flyway, the Mississippi Flyway, the Central Flyway, and the Pacific Flyway (Lincoln, 1935; Jahn and Kabat, 1984; Buhnerkempe et al., 2016). Michigan is located in 2 of these major flyways, the Atlantic and the Mississippi (Lincoln, 1935; Jahn and Kabat, 1984). Alberta is considered part of the Central Flyway, but because the western border of the Mississippi Flyway is not clearly defined, Alberta likely sees migratory waterfowl from the Mississippi Flyway each year (Lincoln, 1935; Jahn and Kabat, 1984).

The first intermediate snail host of the putative novel schistosome is ubiquitous and has a wide geographic range. In North America, *P. trivolvis* has been reported in all of the Canadian provinces and the Yukon territory, as well as across the United States (Clarke, 1981; Lodge et al., 1987; Pip, 1987; Stewart and Dillon, 2004; Stewart, 2006; Johnson et al., 2013). Notably, the genus is closely related to *Biomphalaria*, species of which are known to host schistosome species that cause one of the most devastating helminth diseases that affect humans (Morgan et al., 2002). Even though these snails are considered ubiquitous, researchers have dismissed species of *Planorbella* as hosts for swimmer's itch-causing parasites for almost a century, beginning with Cort in Michigan (Cort, 1928; Cort and Brooks, 1928). During an extensive study of the trematode community in Michigan (Cort and Brooks, 1928), *P. trivolvis* was found not to host schistosome parasites. Moreover, there are no published records of *P. trivolvis* hosting schistosome cercariae until researchers discovered 1 snail hosting a schistosome identified as avian schistosomatid sp. C in Alberta (Gordy et al., 2018). These parasites have been missed by researchers for some time or at least misclassified if they were previously observed. This emphasizes the importance of using sequencing as an identification tool rather than relying on morphology alone.

The snails collected in Michigan during June have a higher infection prevalence with this parasite than those collected in July. In both Alberta and Michigan, *P. trivolvis* snails infected with this schistosome were collected in areas that were conducive to swimming and thus cercarial dermatitis transmission. When collections of *P. trivolvis* snails began in Michigan during the 2018 collection season, it is possible that the peak emergence of this parasite was missed, as collections were not made at the beginning of June in previous years (Blankespoor and Reimink, 1991; Verbrugge et al., 2004; Froelich et al., 2019; Rudko et al., 2019). Furthermore, there was an added objective during the summer 2019 collection season to collect *P. trivolvis* because of the putative novel schistosome discovered in 2018. As such, 2019

Table I. Extended.

Cercariae measurements					
Tail stem (W)	Furcae (L)	Anterior organ (L)	Anterior organ (W)	Snail hosts	References
(23–25)	**(91–98)**	**85.3 (n = 1)**	**55.7 (n = 1)**	***Planorbella trivolvis***	**Present study**
51 (46–57)	221 (219–225)	—	—	*Gyraulus parvus*	Pinto et al., 2017
34 ± 4 (27–48)	222 ± 9 (205–239)	75 ± 11 (55–96)	37 ± 6 (14–48)	*Biomphalaria glabrata*	Pinto et al., 2017
48 ± 4 (42–59)	254 85 ± 13 (67–106) 27 (189–306)	85 ± 13 (67–106)	—	*Anisus leucostoma*	Dönges, 1964
46 ± 3	289 ± 7	71 ± 12	46 ± 4	*Anisus vortex*, *Gyraulus alba*	Aldhoun et al., 2012
29 ± 0.3	135 ± 1.5	—	—	*Gyraulus parvus*	Brackett, 1940
25	150	—	—	*Gyraulus parvus*	Brant and Loker, 2009b
(53–70)	(105–135)	(75–90)	(60–75)	*Planorbarius corneus*	M: Iles, 1949 SH: Iles, 1949; Zbikowska, 2003; Faltýnková et al., 2007
(38–48)	(99–125)	—	—	*Biomphalaria sudanica*	Brant et al., 2006
39.3	124.8	—	—	*Ceratophallus natalensis*	Brant et al., 2006

was much more focused on these snails and is likely the reason that more infected *P. trivolvis* snails were observed.

In the future, efforts will focus on locating the adult worm of this parasite within the definitive host so that a new genus can be proposed and species described. As another future study, the swimmer's itch and cercarial emergence experiments would benefit from more trials. Our swimmer's itch test only had 4 participants that were experimentally exposed to the parasite. To look at this in a more encompassing manner, more volunteers would have to be experimentally exposed to the parasite several times and monitored to record if a rash developed after every exposure. As for the cercarial emergence experiment, further trials would have to be completed with a larger sample size to verify if the emergence during the day is truly sporadic, or if there are peaks.

In summary, the swimmer's-itch problem in Michigan and Alberta has been well documented (Cort, 1928; Jarcho and van Burkalow, 1952; Levy and Folstad, 1969; Blankespoor and Reimink, 1991; Keas and Blankespoor, 1997; Lindblade, 1998; Verbrugge et al., 2004; Gordy et al., 2018; Rudko et al., 2018, 2019; Froelich et al., 2019) and, on the basis of the prevalence of this putative novel schistosome in Michigan, it is likely that it has been an unknown contributor to swimmer's-itch cases in the state for some time. Had we not come across this parasite by chance, a large contributor to the swimmer's-itch problem in Michigan, and perhaps throughout North America, would have gone unnoticed. From now on, surveys of avian schistosomes should consider collecting *P. trivolvis* snails.

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All procedures performed in studies involving human participants were conducted following the ethical standards of the institutional or national research committee and with the 1964 Helsinki Declaration and its later amendments or comparable ethical standards. This research was approved by the University of Alberta Human Research Ethics Board: Approval # Pro00055721. Oral consent was obtained from all survey participants.

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