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Rapid Communication

First record of the parasite *Transversotrema patialense* (Soparkar, 1924) within New Zealand, and its prevalence in *Melanoides tuberculata* (Müller, 1774) among captive and "wild" populations

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Abstract

The trematode parasite *Transversotrema patialense* (Soparkar, 1924) (Digenea: Transversotrematidae) is reported from New Zealand for the first time. The parasite was first observed serendipitously within water surrounding the non-indigenous thiarid snail *Melanoides tuberculata*, a known intermediate host, bought from an online Auckland aquarium trade supplier in April 2016. In 2019, we systematically surveyed *M. tuberculata* from three online suppliers, a home and university laboratory aquaria, and from the only known New Zealand "wild" population, to determine its prevalence. The parasite was found to be present only from snails bought from a single Christchurch supplier. The establishment of *T. patialense* in New Zealand's natural waters will be dependent on the presence of *M. tuberculata*, and appropriate fish hosts, and will therefore likely be limited to geothermally warmed waters. Release of infected snails from aquaria would be required for the establishment of the parasite in the "wild" population, and in other suitable habitats. Further research is necessary to quantify the threat that *T. patialense* poses to native fish in New Zealand, though these are not typically found in geothermally heated waters. The greatest risk of the parasite currently is to the health of captive tropical aquarium fish.

Key words: trematode parasite, Transversotrematidae, cercariae, exotic species, tropical aquaria, biosecurity

Introduction

A common source of parasite invasions is via infected hosts being introduced to new habitats (Prenter et al. 2004). There is then the risk of these non-native parasites infecting new native hosts via parasite spill-over (Daszak 2000). A trematode parasite associated with the thiarid snail *Melanoides tuberculata* (Müller, 1774), purchased from an Auckland, New Zealand, online aquarium supplier, was observed in April 2016. Here, we identify this as the non-indigenous parasite *Transversotrema patialense* (Soparkar, 1924) (Digenea: Transversotrematidae). *Transversotrema patialense* relies on two host stages through its life cycle; known intermediate hosts comprise only the thiarid snails *Sermyla riquettii* (Grateloup, 1840) and

three species of *Melanoides*, with most records from *M. tuberculata* (Womble et al. 2015). Having only two hosts, rather than the typical three, makes transversotrematids unusual among trematodes (Poulin and Cribb 2002); this relative lifecycle simplicity may increase the probability of establishment of species in this family compared to other trematodes. *Melanoides tuberculata* has a broad distribution, having been introduced widely, but originates from the Middle East and East Africa (Pointier 1999). This snail represents the only known intermediate host of *T. patialense* present in New Zealand (Spencer et al. 2009). The only known "wild" population of this snail in New Zealand is at the geothermally heated Golden Springs, near Taupō (Duggan 2002). This species is also a common inhabitant of tropical aquaria in New Zealand, where it is commonly considered a pest, introduced in conjunction with aquatic plants (Duggan 2010). However, sellers on online trading sites advertise these snails as being useful for consuming algae, and as food for snail-eating fish such as *Botia* (Indian loaches). Definitive hosts of *T. patialense* include a wide variety of freshwater and euryhaline fish species, including *Danio rerio*, *Scatophagus argus* and *Planiliza subviridis* (Whitfield and Wells 1973; Womble et al. 2015). *Transversotrema patialense* is distributed widely, known from Africa, India, Sri Lanka, Malaysia and Australia (Cribb et al. 1992). This study documents *Transversotrema patialense* for the first time in New Zealand, discusses the potential impacts it may have, and considers possible methods of dispersal. We also examined the prevalence of the parasite among *M. tuberculata* obtained from various suppliers, home and laboratory aquaria, and the "wild" population at Golden Springs.

Materials and methods

For the systematic survey of parasite prevalence, specimens of *M. tuberculata* were purchased from one Auckland, one Wellington, and one Christchurch supplier via the New Zealand online auction website TradeMe during March 2019; these were typically advertised in lots of > 10 individuals per sale. Snails obtained from a University of Waikato (Hamilton, N.Z.) laboratory aquarium in March 2019 and a Hamilton home aquarium in April 2019 were also examined. On 16 April 2019, *M. tuberculata* were collected from the "wild" population at Golden Springs (38°28′12″S; 176°18′38″E). Snails were observed visually and obtained from the silty margins of the stream by hand.

After the arrival of each sample at the laboratory, the water in which the snails were contained was examined in a Petri dish under an Olympus SZ60 stereoscope for *T. patialense* cercariae. During transportation, snails were subjected to prolonged periods of darkness due to their packaging, which could induce cercaria shedding (Whitfield et al. 1977). If parasites were not identified after initial examination of the transport water, then snails

Table 1. Number, maximum, minimum, and mean lengths (mm), and standard deviation of *Melanoides tuberculata* from each source and presence of trematode parasite *Transversotrema patialense*.

	Golden Springs "wild" population	Laboratory aquarium, Hamilton	Home aquarium, Hamilton	Auckland supplier	Wellington supplier	Christchurch supplier
Number examined	129	10	21	66	16	18
Maximum length (mm)	22.6	8.5	16.9	15.1	11.6	14.6
Minimum length (mm)	6.1	5.8	11.5	6.7	7.7	11.0
Mean length (mm)	13.3	7.1	14.0	10.7	9.5	13.1
Standard deviation	3.0	0.9	1.4	1.9	1.2	1.1
Presence of *T. patialense*	Absent	Absent	Absent	Absent	Absent	Present

were placed in a dark temperature-controlled cabinet set at 26 °C in Petri dishes containing aged tap water, following Whitfield et al. (1977). Samples were examined daily for up to four days before *T. patialense* parasites were deemed to be absent. All snail specimens were measured using an electronic Vernier Calliper Gauge. Preserved cercariae, obtained from the online Auckland aquarium trade supplier purchased in April 2016, were also re-examined to confirm their identity. *Transversotrema patialense* specimens were mounted under a coverslip on a slide in glycerol, and photographed using a Nikon SMZ25 microscope.

Results

Results obtained from snails collected from the systematic survey are provided in Table 1, including the number of snails collected, maximum, minimum, mean lengths and standard deviations, and the presence of *T. patialense*. The largest number of *M. tuberculata* individuals were collected from the Golden Springs population, which were divided into three groups during incubation. Of the six different populations of *M. tuberculata* sampled during the present study, only one was found to have the target parasite *T. patialense* present. The sample purchased from the Christchurch online supplier was found, upon first examination following delivery, to contain multiple cercariae swimming within the water in which the snails were transported. The distinctive parasites were identified using the review of the family Transversotrematidae by Cribb et al. (1992). This family is distinctive in the transversely elongate shape of the adults, and as the cercariae resemble completely developed adults' forms, with a furcate tail and distinctive arm processes at their bases (Cribb et al. 1992). In this review, several morphologically similar freshwater species that utilise *M. tuberculata* as an intermediate host were synonymised as *T. patialense* (*T. koliensis, T. laruei, T. chackai* and *T. soparkari*). Comparison of cercaria obtained in the current study with *T. patialense* presented by Cribb et al. (1992) and Ben-Ami et al. (2005) shows the characteristic shape and appendages of this species (Figure 1). Re-examination of the Auckland specimens, bought in 2016, showed them to be identical.

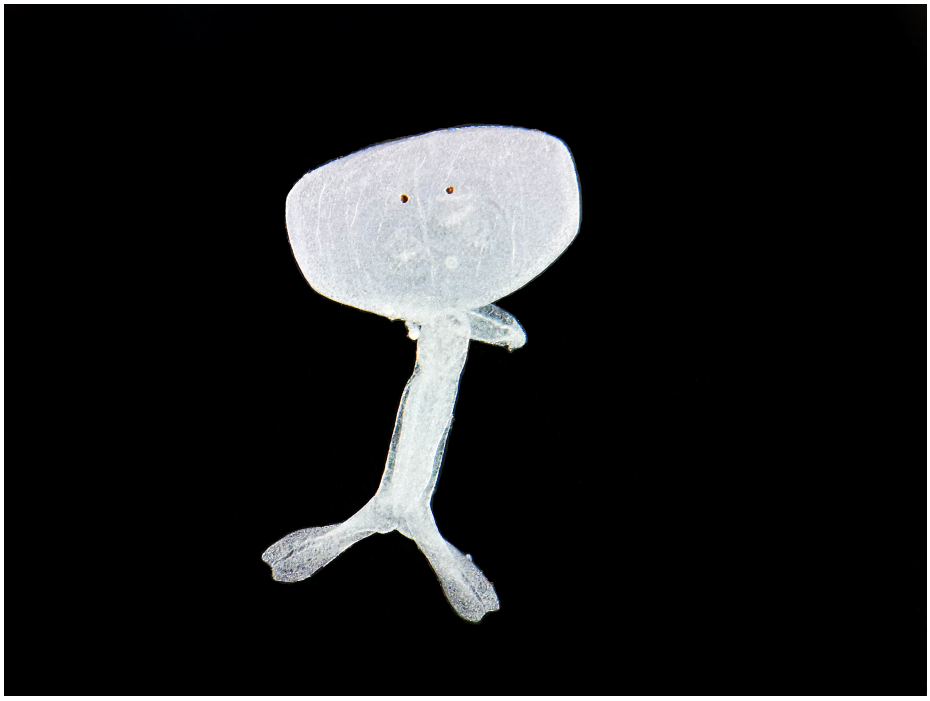

Figure 1. Dark field image of a *Transversotrema patialense* cercaria shed by *Melanoides tuberculata* originating from an online Christchurch supplier. Photo: Nigel Binks, University of Waikato.

Discussion

We identified the cercariae collected from an Auckland commercial supplier in April 2016 (not resampled here) as *T. patialense* and recorded further individuals from snails from a Christchurch supplier. None of the other sources examined in the present study had *T. patialense* present. The morphology of the specimens conformed to *T. patialense* and is unlikely to be confused with the cercariae of any other species. Nevertheless, while the morphology of *T. patialense* is distinctive, and the species has a limited number of snail hosts, due to the synonymisation of several species of freshwater *Transversotrema* by Cribb et al. (1992), we recommend molecular markers be used to determine the diversity of this genus. Currently, however, no sequences of *T. patialense* are available on Genbank.

Transversotrema patialense is a highly specific parasite regarding its intermediate hosts, whereas it is known to infect a wide variety of fish species as definitive hosts, although both are required in order to complete its life cycle (Whitfield and Wells 1973). The occurrence of *T. patialense* in New Zealand is dependent firstly on the presence of *Melanoides tuberculata,* its most commonly reported intermediate host elsewhere. In order for this parasite to have established within the suppliers' *M. tuberculata* populations, there must also be suitable definitive hosts living within the same space, such as susceptible tropical fish (e.g. Poecilidae species) (Womble et al. 2015). Recent importation of infected *M. tuberculata* from overseas is unlikely, as the most likely vector for movement of the snails—aquatic macrophytes—are almost exclusively domestically cultured in New

Zealand (Duggan et al. 2018). A possibility exists that infected snails originated from the domestic suppliers of aquatic plants. Sale of infected snails by suppliers may facilitate the spread of *T. patialense* populations among aquaria in New Zealand (Keller and Lodge 2007; Derraik and Phillips 2010), though our records indicate that to date infections among aquaria do not appear to be widespread. Nevertheless, it is possible that the presence of *T. patialense* may have been underestimated among our *M. tuberculata* samples, as snails harbouring prepatent infections may fail to release cercariae. We therefore recommend dissection to more confidentially determine the prevalence of the parasite in future studies.

The aquarium trade is known to be a source of non-indigenous invertebrates, including *M. tuberculata* (Duggan 2010; Ng et al. 2016; Duggan et al. 2018). *Melanoides tuberculata* occurs in the "wild" in New Zealand at just one location, Golden Springs (Duggan 2002). Minimum tolerable temperatures of *M. tuberculata* have been reported as 18 °C to 21 °C (Murray 1971; Ismail and Arif 1993), and their survival in New Zealand natural waters is thus dependent on them being geothermally heated (Duggan 2002). *Melanoides* at Golden Springs may have been introduced as early as the 1920s and were possibly released in concert with the guppy (*Poecilia reticulata*), which is known there (McDowall 1990; Duggan 2002). Interestingly, three species from the same family as the guppy (Poecilidae) are known to be hosts of *T. patialense* (Velasquez 1961; Wongsawad et al. 2004; Ben-Ami et al. 2005; Womble et al. 2015). This includes sailfin molly (*Poecilia latipinna*) and the mosquitofish (*Gambusia affinis*), both of which are also present in New Zealand, with the sailfin molly occupying thermally warmed waters suitable for *M. tuberculata* (McDowall 1990). The presence of these known definitive hosts could be of concern if *M. tuberculata* were to become more widely distributed or if snails are introduced where fish are present. The possibility of *P. reticulata* also being susceptible means that both an intermediate and definitive host may occupy the same habitat, meaning an incursion of *T. patialense* at Golden Springs could allow the parasite population to establish there. Experimental determination of whether guppy can act as an appropriate host is warranted. Nevertheless, dispersal of infected *M. tuberculata* to suitable habitats within the Taupō Volcanic Zone, and around New Zealand, is unlikely to occur without human influence due to the limitations of distance and temperature (Duggan 2002).

The sizes of *M. tuberculata* measured were similar in the parasitized population as the other captive populations. It has been reported elsewhere that parasitized *M. tuberculata* may exhibit gigantism (Minchella 1985). Interestingly, the snails from the unparasitized Golden Springs population had some individuals much larger than those found in the captive samples; the range and standard deviation of this "wild" population were also greater.

Introduced parasites can have negative effects on native communities via parasite spill-over, and the establishment of *M. tuberculata* in New Zealand may expose *T. patialense* to naïve hosts with no effective defence mechanisms (Daszak 2000; Prenter et al. 2004). Nevertheless, a lack of native fish species living in geothermal streams limits this possibility. The health of aquarium fish may thus represent the greatest risk of the presence of *T. patialense* in New Zealand. Suitable hosts occupying a limited space with infected snails may expose fish to high parasite loads. *Melanoides tuberculata* are often considered a contaminant in aquaria, and effort should be made by aquarists to remove the snails from tanks in case they are parasitized. However, the ways in which digenean parasites affect their definitive hosts are still poorly known (Eiras et al. 2008). A concerning aspect of *T. patialense* being present in New Zealand is the possibility of other trematode parasites also establishing. For example, a number of trematodes that utilize *M. tuberculata* as an intermediate host are a threat to human health (Derraik 2008). Attention is required to quantify both the threat of *T. patialense* being present in New Zealand and of other trematode parasites potentially establishing in the future.

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