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Short Communication

Melanoides tuberculata as intermediate host of *Centrocestus formosanus* (Nishigori, 1924) in Tunisia

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Melanoides tuberculata that naturally harbored trematode larvae were collected at the irrigation system in Ain Soltan's oasis (southwest of Tunisia), during malacological surveys conducted from January to December 2013. From 2160 specimens of *M. tuberculata* collected, 31 (1.4%) were infected by the cercariae of *Centrocestus formosanus* (Digenea: Heterophyidae). The parasite was present in snails larger than 19 mm. This is the first report of *C. formosanus* in *M. tuberculata* in Tunisia.

Key words: Centrocestus formosanus, Melanoides tuberculata, Tunisia, intermediate host.

INTRODUCTION

Snail species are the first intermediate host for most of parasitic trematodes that provoke diseases such as heterophyidiasis, schistosomiasis, and fascioliasis. Melanoides tuberculata (Müller, 1774) is a fresh water snail, which lives in the bottom of water and in abundance in the irrigation system in Ain Soltan's oasis in the south west of Tunisia. It has been recorded as the first intermediate host of the tremadote Echinochasmus japonicus, which is a trematode of birds and mammals, and belongs to the Echinostomatidae. (Cheng and Fang, 1989), which occasionally may infect human beings (Sayasone et al., 2009). Furthermore, M. tuberculata also harbors the trematode Clonorchis scinensis, agent of the clonorchiasis disease (Kino al., 1998). et Xiphidiocercariae, aquatic larval stages of some trematodes are considered a potential instrument for biological control of mosquitoes. This tramatodes have been described from seven Egyptian snails, including *M. tuberculata* collected from Giza and Qualiobyia (Wanas et al., 1993). The aim of this study was to determine the cercarial fauna of *M. tuberculata* snail and their role in transmitting diseases in Ain Soltan's oasis (south-western Tunisia).

MATERIALS AND METHODS

M. tuberculata snails were collected manually from irrigation system in the oasis of Ain Soltan in the governorate of Gafsa, Tunisia (34°27'13"N 8°47'44"E) from January to December 2013 to find the cercariae,.

Collected snails were transferred in plastic bags to the laboratory, where they were thoroughly cleaned and maintained for several

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Figure 1. *Melanoides tuberculata* snail from Ain Soltan's oasis of Southwestern Tunisia.



Figure 2. The cercariae of *Centrocestus formosanus.*

weeks in aquaria with a thin substrate of sand and dechlorinated water and provided with green algae for feeding.

They were counted, measured using caliper and examined for cercariae by crushing method. In this method, shell of snails was

broken and soft tissues were placed between two slides then squashed. Collected cercariae were studied live, unstained and stained with vital stains, and fixed in 5% formalin, preserved in 70% ethanol, cleared in lactophenol and stained with a 10% Rose Bengal solution. Body proportions were measured by ocular micrometer.

RESULTS

In all, 2160 specimens of *M. tuberculata* (Figure 1) were collected and examined. Cercaria emerged from 31 *M. tuberculata* specimens (1.4%). Cercaria was preliminarily characterized as belonging to the family Heterophyidae, genus *Centrocestus* (Figure 2). Cercariae tended to emerge during the morning, in small numbers, and were very active.

Cercariae presented Piriform body, morphologically agreeing with the descriptions and drawings of *C. formosanus* from *M. tuberculata* of China, Hong Kong, Taiwan, and Mexico (Chen, 1948; Arizmedi, 1992), and *S. newcombi* of Hawaii (Martin, 1958).

Measurements (given in μ m, with average in parentheses) of 20 fixed and stained specimens, without cover glass pressure, were: body length of 114-156 (141); body width of 77-104 (84); oral sucker length of 29-38 (35); oral sucker width of 26-34 (39); pharynx diameter of 13-16 (14); ventral sucker diameter 20-25 (23); tail length of 115-156 (132); tail width pf 16-22 (18).

Under test conditions, the cercariae of *C. formosanus* swam in more or less straight lines. *M. tuberculata* whose size shell length is less than 19 mm showed no larval forms of *C. formosanus*. The parasite is only present in snails larger than 19 mm. Infection rate increases exponentially with snail body size (Table 1 and Figure 3). The regression fit is:

Infection rate = 0.107 - 7.523*Shell size.

The correlation coefficient was R = 0.999 and this function explains 99.984% of variance in infection rate.

DISCUSSION

The life cycle of *C. formosanus* has primarily been elucidated in Taiwan, where the developmental stages and the participation of thiarids mollusks in its transmission have been described (Nishigori, 1924). Since then, several studies have confirmed the participation of *M. tuberculata* as an intermediate host of *C. formosanus* in different countries, such as China (Chen, 1948; Chao et al., 1993), Japan (Yanohara, 1985; Yanohara, et al., 1987), Mexico (Salgado-Maldonado et al., 1995; Scholz et al., 2000; Scholz and Salgado-Maldonado, 2000), Malaysia (Bayssade-Dufour et al., 1982), Taiwan (Lo and Lee, 1996a, b), India (Madhavi et al., 1997), United States (Tolley-Jordan and Owen, 2008), Venezuela (Hernandez et al., 2003), Iran

Table 1. Variation of infection rates according the size of *Melanoides tuberculata*.

Size of snail (mm)	<3	<10	<15	<20	<25	<30	<35	<40	Total
Snail samples	248	415	353	304	629	169	37	5	2160
Snail samples infected	0	0	0	0	8	10	8	5	31
Infection rate (%)	0	0	0	0	1.3	5.9	21.6	100	1.4



Figure 3. Infection rate = f (Shell size).

(Farahnak et al., 2005), Colombia (Velásquez et al., 2006; Vergara and Velásquez, 2009) and Brazil (Pinto and Melo, 2010). Despite these reports, the possible involvement of *M. tuberculata* in the transmission of *C. formosanus* has still not been reported in Tunisia.

In the present study, the participation of *M. tuberculata* in the biological cycle of *C. formosanus* in Tunisia is confirmed, but the natural definitive host of *C. formosanus* in Ain Soltan's oasis remains unknown. Knowing that there are freshwater fish and birds associated to *M. tuberculata*. Given that this snail is widespread in Tunisia and participates in the life cycle of *Centrocestus* in the country, the impact of introducing and spreading these thiarids around the country must be better evaluated, with an aim toward preventing future cases of Centrocestiasis. Centrocestiasis is a gill trematode disease of numerous cultured and wild fish species that is caused by a digenetic trematode *Centrocestus formosanus*.

In this study, only snails larger than 19 mm are infested, implying that the infestation rate increases with increase in the size of *M. tuberculata*. Also, in Texas the parasite is usually only present in snails larger than 17 mm (Mitchell et al., 2000).

Conflict of Interests

The author(s) have not declared any conflict of interests.

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