Egg predation and parasite prevalence in the invasive freshwater snail, *Melanoides tuberculata* (Müller, 1774) in a west Texas spring system

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**Abstract**

*Melanoides tuberculata* Müller, 1774 (Thiaridae), a freshwater prosobranch snail native to regions of Asia and Africa, was introduced into the U.S. in the 1960s and is now found in fifteen states. *Melanoides tuberculata* can affect native communities directly by displacing native snail species and indirectly by introducing foreign trematodes into novel environments. As the exact mechanisms of displacement of native snails by *M. tuberculata* are unknown, egg predation rates on native snails (*Physella* spp.) by two different size classes (>30 mm and <30 mm, total length) of *M. tuberculata* were determined in laboratory experiments. Additionally, *M. tuberculata* were sampled from Diamond Y Spring (Pecos County, TX) to determine if the exotic trematode *Centrocestus formosanus* (Nishigori, 1924) has been introduced into the system. No *M. tuberculata* > 30 mm were found to consume *Physella* spp. and only 2.8% of *M. tuberculata* snails consumed egg masses. None of the *M. tuberculata* collected from Diamond Y Spring were found to be infected with *C. formosanus*.

**Key words:** *Centrocestus formosanus*, Diamond Y Spring, *Melanoides tuberculata*, predation, Texas

**Introduction**

Invasions of ecosystems by non-native species are increasing, especially in aquatic environments (Byers 2000). The consequences of these invasions include biodiversity loss and changes in ecosystem structure (Pimentel 2002). Specifically, the establishment of non-native freshwater snails has been associated with changes in ecosystem function (Arango et al. 2009), community structure (Kerans et al. 2005), and the introduction of non-native parasites (Madsen and Frandsen 1989). *Melanoides tuberculata* Müller, 1774 (Prosobranchia: Thiaridae), a snail native to regions of Asia and Africa (Brown 1980), has been confirmed in at least 15 states throughout the U.S., including California, Texas, Arizona, Louisiana, and Oregon (Karatayev et al. 2009). Thiarid snails, such as *M. tuberculata*, can directly alter community structure through competition with native snails (Pointer et al. 1993). In fact, *M. tuberculata* has been introduced in areas where schistosomiasis was prevalent in the hopes of reducing populations of vector snails such as *Biomphalaria glabrata* (Say, 1818) and *B. straminea* (Dunker, 1848), and thereby reducing the incidence of the disease (Pointer et al. 1984; Pointer and Guyard 1992). Yet, quantitative analyses describing the interactions of *M. tuberculata* with native snails are lacking (Tolley-Jordan and Owen 2008) and it is not known whether native snails are displaced as a result of competition, displacement, or perhaps even predation. Competition and intraguild predation has been shown in some freshwater snail species (Turner et al. 2007). *Melanoides tuberculata* prey upon the eggs of endangered fishes (Phillips et al. 2010) so it is conceivable that they may also prey on the eggs or juveniles of other snails. However, *M. tuberculata* are generalist grazers and the ingestion of eggs may not be intentional, but an indirect effect of grazing.

Another major concern with the introduction of exotic snails is the potential parasites that they may carry and their effects on native fauna. Non-native trematodes have been documented to
negatively affect native communities by infecting vertebrate hosts lacking the co-evolved defense mechanisms to keep densities of parasites at a low level (Taraschewski 2006). Melanoides tuberculata serves as the first intermediate host to several species of digenetic trematodes, including the gill trematode Centrocestus formosanus (Nishigori, 1924) (Ben-Ami and Heller 2005; Tolley-Jordan and Owen 2008). Centrocestus formosanus infects some vertebrate hosts, such as aquatic migratory birds (e.g. the green heron Butorides virescens (Linnaeus, 1758) and great egret Ardea alba Linnaeus, 1758) (Mitchell et al. 2005) as well as a wide variety of fish hosts, including several endangered spring species [e.g. the fountain darter, Etheostoma fonticola (Jordan and Gilbert, 1886)] (Mitchell et al. 2000). The presence of C. formosanus produces inflammatory responses in the gills of many freshwater fishes that can lead to respiratory stress and eventual mortality (Mitchell et al. 2000; 2005).

One of the largest and last remaining cienega systems in west Texas is the Diamond Y Spring preserve located in Pecos County, Texas. It is a 1502-acre tract of land owned and operated by the Nature Conservancy (Karges 2003). The Diamond Y Spring preserve is composed of a number of springs, seeps, and wetlands. The saline waters within the preserve provide critically important habitat for two federally endangered species of desert fishes (Cyprinodon bottinis Baird and Girard, 1853 and Gambusia nobilis (Baird and Girard, 1853)) and a suite of rare aquatic invertebrates. The non-native M. tuberculata is well established within the Diamond Y Spring preserve, albeit no snails have previously been documented to harbor any exotic trematodes (McDermott 2000). However, the spread of C. formosanus into the preserve is of major concern due to its ability to compromise the health of the preserve’s federally endangered fish species. Additionally, it is not known how M. tuberculata affects Diamond Y’s invertebrate fauna. Investigating the effects of M. tuberculata on the preserve’s species of high conservation concern is necessary to effectively manage these rare native populations. Thus, the aims of this study were to determine whether M. tuberculata prey upon the eggs of a native snail (Physella spp.) and if the exotic trematode, C. formosanus, is present in the M. tuberculata population within the Diamond Y Spring preserve.

### Materials and methods

#### Egg predation

The Diamond Y Spring preserve contains two endemic springsnails [Tryonia circumstriata (Leonard and Ho, 1960) and Pseudotryonia adamantia Taylor, 1987], two native snails (Physella spp. and Assiminea pecos Taylor, 1987), and the invasive M. tuberculata (Ladd 2010). The Physella spp. was used for our egg predation trials as they are common and readily produce egg masses year round in a laboratory setting (personal observation). Also, the endemic springsnails are state threatened, while A. pecos is federally listed as endangered and little is known about their reproduction or maintaining them in captivity. Melanoides tuberculata appeared to considerably vary by size within Diamond Y, the only spring where this species is found. No snails ≥ 30 mm were found beyond 300 m downstream of the immediate outflow of the spring head (Ladd 2010). Therefore, we chose to use two size classes in this experiment (>30 mm or <30 mm). The experiment was conducted from June-September of 2010 at Texas Tech University (Lubbock, TX). Adult Physella spp. were stocked into three polypropylene “shoebox” containers (28×14×11 cm deep) filled with de-chlorinated tap water. Snail egg masses were then placed within each vial and predation was monitored for five days. We considered an egg mass to be consumed if the entire egg mass was gone at the end of the trial. A total of 51 M. tuberculata >30 mm and 56 M. tuberculata <30 mm were used over the duration of the experiment. Snails were not reused or fed during the experiment, nor was the water changed.

#### Snail infection status

Of the six snail species found within Diamond Y Spring preserve, only M. tuberculata is known to serve as an intermediate host for C. formosanus (Scholz and Salgado-Maldonado 2001; Mitchell et al. 2005). Therefore, only M. tuberculata were surveyed for the presence of C. formosanus. Live M. tuberculata were collected during October and November of 2009 and March, April, and May of 2010 from Diamond Y spring. Only snails > 17 mm were used during this study since that is the minimum length recorded for infection.
in *M. tuberculata* in the Comal River, Texas (Tolley-Jordan and Owen 2008). Presence of trematodes was assessed using two methods. The first method consisted of cracking the shell of *M. tuberculata* between the first and second body whorl, removing the digestive tract and gonads, and examining each for rediae and cercaria of *C. formosanus* under a binocular dissecting microscope at 100× total magnification. The second method consisted of placing individual snails into 50 ml vials filled with 35 ml of de-chlorinated water and setting two lamps equipped with 60-watt incandescent light bulbs over the vials for 24 hours to ensure that the temperature of the water reached at least 28°C (Lo and Lee 1996). We then pipetted two ml of water from each vial into petri dishes and looked for swimming cercaria using a dissecting scope at 30–40× total magnification. A total of 250 snails were examined for parasite presence. Fifty and 200 snails were examined using the first and second method, respectively. The second method was adopted as our preferred means of assessment as it has been found to be a quick and effective way of determining snail infections [*C. formosanus* cercariae emerge from infected snails when exposed to light and temperatures between 15–35°C (Lo and Lee 1996)].

As parasites in hosts can be in low numbers and patchily distributed (Anderson and May 1979), we wanted to determine if our sample size of 250 was adequate for detecting infected snails. A Monte Carlo simulation was used to determine the probability of obtaining a sample of 250 snails from a population with trematode prevalence levels of 1–5% and observing no infected snails within the sample. Using PopTools® (Hood 2010), we simulated a population of 1000 snails with a 5% infection rate. Two hundred and fifty snails were randomly sampled from that population 4999 times. Then, we counted how many times the sample of 4999 equaled zero infected snails. We repeated this procedure for 4, 3, 2, and 1 percent parasite prevalence.

### Results and discussion

None of the *M. tuberculata* >30 mm consumed any egg masses, while only 3/56 (5.4%) egg masses were consumed in trials using *M. tuberculata* <30 mm. Of 250 *M. tuberculata* collected from the spring, none were found to be infected with *C. formosanus*. Results of the simulation indicated that the probability of obtaining zero parasitized snails in a sample of 250 snails at 5, 4, 3, 2, and 1 percent parasite prevalence was 0.000, 0.000, 0.008, 0.0776, and 0.0826, respectively.

Though the total percentage of egg predation in our experiment was 2.8% (3/107), *M. tuberculata* may be present in large enough numbers to affect native snail populations directly through egg predation. For example, the estimated abundance of *M. tuberculata* within Diamond Y Spring is 187,000 individuals (Ladd 2010). If only 2.8% of *M. tuberculata* consume egg masses, approximately 5000 snails could conceivably consume native egg masses, which may lead to a significant reduction in native snail populations. Future studies focusing on food preference, egg consumption, and natural egg mortality rates *in situ* are necessary to understand the significance of egg predation as a mechanism for displacing native snail species by *M. tuberculata*.

Results from our study and a previous study conducted by McDermott (2000) indicate that the trematode *C. formosanus* is not likely present at the Diamond Y Spring preserve. In *M. tuberculata* native habitats, trematode prevalence is less than 5%, and infections tend to be spatially aggregated (Anderson and May 1979). However, in areas where the snails have been introduced, infection rates can be as high as 53% (Bogéa et al. 2005). Although we did not detect any trematodes, this does not mean that they are not present within the *M. tuberculata* population of the Diamond Y Spring preserve. According to the results of our simulation, parasite prevalence could be two percent or lower.

A variety of fish hosts and three species that serve as definitive hosts for *C. formosanus* have been documented at the Diamond Y Spring preserve: the green heron (*B. virescens*), great egret (*A. alba*), and black-crowned-night heron (*N. nycticorax*; Linnaeus, 1758) (Mitchell et al. 2005; J. Karges, personal communication). In addition, *C. formosanus* has been documented in fishes and *M. tuberculata* from other west Texas springs (McDermott 2000), which suggests that fishes and *M. tuberculata* from Diamond Y Spring are likely to become infected, eventually. Therefore, we strongly recommend yearly sampling of *M. tuberculata* for *C. formosanus* within Diamond Y Spring and other west Texas springs to monitor the spread of this harmful fish pathogen.
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