

Investigating *Echinostoma trivolvis*

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Echinostoma trivolvis

Diversity

The phylum Platyhelminthes is divided up into four different classes, including Trematoda. Trematodes are further divided into more groups that include the Digenea and Monogenea, with digenetic trematodes totaling over 24,000 species (Poulin & Morand, 2004). The monogeneans were initially confused as part of the digenetic trematodes but were found to differ in the methods of parasitizing the host (Monogenea are exoparasites while Digenea are endoparasites) and the lack of an intermediate host (Digenea infect an intermediate mollusk host). In addition, Monogenea are typically parasitic to fish and not mammals or birds, like members of Digenea (Cribb et al., 2002) *Echinostoma trivolvis* is one of roughly 100 species in its genus. Like most other trematodes, *E. trivolvis* is unsegmented, has a pair of suckers, an incomplete digestive system, a primitive brain, and protonephridia (flame cells) that aid in removing waste, and it is hermaphroditic (individuals have both female and male sex organs) (Bush et al., 2001).

Systematic and Taxonomic History

Parasites are usually classified by their morphology and the host(s) they parasitize. However, many parasites species are difficult to identify or distinguish, since they often exhibit simplified morphology, parasitize similar hosts, and exhibit wildly different morphologies throughout their life cycle. The difficulty of culturing a parasite through several host organisms also makes taxonomy problematic. *Echinostoma trivolvis* was initially confused with *Echinostoma revolutum*. Many past scientific articles studied *Echinostoma trivolvis*, but identified it incorrectly as *Echinostoma revolutum*. For example, there were over 60 studies conducted by B. Fried and his colleagues on “*E. revolutum*” between 1968 and 1988, in addition to articles written by Paul C. Beaver in 1936 and Clyde M. Senger in 1954 (Baker & Muller, 1996). Fried and colleagues also were the ones to correct the identification (Kanev 1995). *E. trivolvis* differs from most other trematodes because of its 37-collar-spine and definitive hosts, which can be a bird or muskrat (Mucha et al., 1990). When compared to *E. revolutum*, the adult stage of *E. trivolvis* can infect both birds and mammals, while *E. revolutum* adults only infect birds. *E. trivolvis* is also distinguished by cercariae that only infect Planorbid snails; *E. revolutum* infect Lymnaeid snails. *E. trivolvis* occur across North America, while *E. revolutum* is found in Europe and Asia (Kanev, 1995).

Physical Description

The anatomy of *Echinostoma trivolvis* changes dramatically through its life cycle. As an adult, *E. trivolvis* is a 37-collar-spined trematode (Fried et al., 1997). The 37-collar-spines refer to the spines found around the mouth of the trematode, attached to the distal cytoplasm (Roberts & Janovy Jr., 2000). The distal cytoplasm is another name for a trematode’s tegument, a secreted cuticle used for protection and defense against water loss. Like other species of the Phylum Platyhelminthes, *Echinostoma trivolvis* lacks circulatory, respiratory, and skeletal systems (Bush

et al. 2001). They have protonephridium (also known as flame cells) that aid in the excretory system, similar to the function of human kidneys.

Both *Echinostoma* eggs and miracidia are roughly 100 micrometers long, while Digenea adults vary in size, but typically range from 1 to 5 mm (Fried & Graczyk, 1997). Miracidia have cilia that allow for movement to find a host, while cercariae have a flagellum (Bush et al., 2001).

Echinostoma trivolvis metacercarial cysts are spherical with walls partially created from pieces of the host's kidneys (Fried & Bennett, 1979). Trematodes, like *E. trivolvis*, have both male and female reproductive organs. The male part typically has two testes, while the female region has one ovary (Fried & Graczyk, 1997).

Development

The lifecycle of *Echinostoma trivolvis* consists of seven basic stages (Fig 7), which include two transmission or infective stages, miracidia and cercariae (Davis, 2005).

- **Eggs** (Fig. 1) are produced, as a result of sexual reproduction by hermaphroditic adult worms within the definitive host. They are released with the host's excrement and take approximately 2-3 weeks to develop into **miracidia** (Fig. 2) in an aquatic environment (Baker & Muller, 1996).



Figure 1 Egg

- The miracidia find and infect a first intermediate host, the gastropod *Planorbella (Helisoma) trivolvis*, by penetrating the head-foot area of the snail (Toledo et al., 2007). Penetration takes roughly 30 minutes for most trematodes (Roberts & Janovy Jr., 2000).



Figure 2 Miracidia

- Inside the first host, the miracidia form into a sac, called a **sporocyst**. Embryos form and develop into **redia** (Fig. 3) and into daughter redia via asexual reproduction. These, in turn, can produce many **cercariae** (Fig. 4) through asexual reproduction and exit through a birth pore (Detwiler & Minchella, 2009).

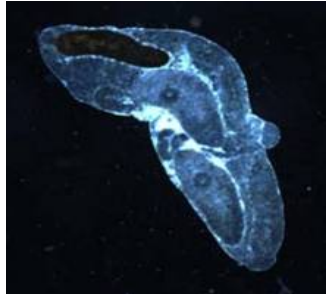


Figure 3 Redia



Figure 4 Cercariae

- After 4-6 weeks in the gastropod, the cercariae leave the first host (Baker and Muller, 1996). They can then infect a number of different second intermediate hosts, including both gastropods and amphibians. In this second intermediate host, the trematode develops into a **metacercaria** (Fig. 5) that will encyst, typically in the kidneys, for several weeks to years (Detwiler & Minchella, 2009).

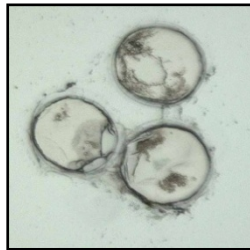


Figure 5 Metacercariae

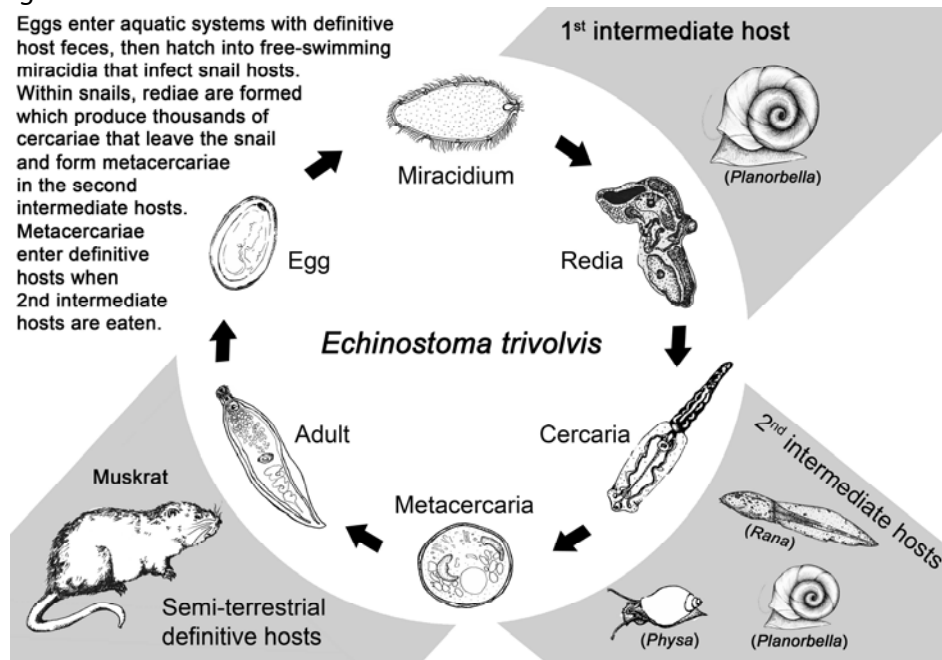
- Metacercariae eventually enter the definitive host via consumption of the second intermediate host. The definitive host, either a bird or muskrat, will become the habitat for the **adult** *Echinostoma trivolvis*. In a survey of possible definitive hosts in SW Virginia, no adult *E. trivolvis* were found in geese while 15/21 necropsied muskrats had adult *Echinostoma*, with a range of 1-113 worms/muskrat and a median of 49 worms (L. Belden, *unpublished data*). The adult parasite will most likely remain in the digestive tract, eventually laying eggs that will finally enter an aquatic habitat, starting the lifecycle over again. (Bush et al., 2001).



Figure 6 Adult

Studies conducted by N. E. Davis investigating the eggs of *Echinostoma revolutum*, a closely related species, have shown that the duration of hatching time in trematodes depend the temperature of the environment. Eggs can remain dormant until warm water temperature to avoid the chance of freezing as miracidia. The ideal water temperatures for these parasites vary depending on the habitat or if they are in controlled laboratory settings. In addition, temperature also affects the miracidia and cercariae stages. As temperature is increased, activity increases, which causes the parasite to use more resources from the host (Davis, 2005).

Figure 7.



Photos/Life cycle diagram: Dr. Lisa K. Belden, Virginia Tech.

Geographic Range

Geographic range and population size of a trematode is usually dependent on host availability and movement in nature (Detwiler & Minchella, 2009). On the continental scale, *E. trivolvis* are found only in North America (Kanev, 1995; Klockars et al. 2007). Their first intermediate hosts, *Planorbella trivolvis*, has a widespread distribution across the continent, only absent from the northern Canadian provinces, the Pacific northwest, the desert southwest, and perhaps several great plains states (Burch 1989). Their primary definitive hosts, the common muskrat *Ondatra zibethicus*, are distributed across the entirety of North America, excepting the northernmost parts of Canada, the desert southwest and parts of California (Patterson et al., 2003). From these first-intermediate and definitive host distributions, coupled with *E. trivolvis*'s low second-intermediate host-specificity, it is tenable to suggest a very wide distribution of *E. trivolvis* across North America. At the local scale, Klockars and her associates (2007) studied sites in

New Jersey with *Helisoma trivolvis*, the first intermediate host, to determine the prevalence of parasites found in the organism. Nine of ten sites studied were found to have *H. trivolvis* infected with *E. trivolvis*.

Habitat

For organisms with complex life cycles, including several free-swimming stages and several hosts, defining the organism's habitat is difficult. The habitat of the parasitic stages is primarily dictated by parasite location inside hosts (Bush et al., 2001). For example, as an adult, *E. trivolvis* inhabit the digestive system of vertebrates – but even within the intestines of a muskrat there is a wide range of conditions along the length of the intestine (i.e. changes in pH). Most often scientists' understanding of the physical requirements of a parasite, in terms of within-host habitats, is poorly known. Habitat also changes ontogenetically, that is, as the parasite go through various stages in its lifecycle. In the first host, snails are infected in the ovotestis and hepatopancreas, an organ that functions like the liver and pancreas in mammals, by miracidia (Fried et al., 1987). *Echinostoma trivolvis* metacercariae encyst in the kidneys of snails and amphibians (Schmidt & Fried, 1996), while most adults live in the digestive tract of the definitive host, which can be a muskrat or bird (Fried & Graczyk, 1997).

Food Habits

Trematodes, like *E. trivolvis*, are facultative anaerobes (Roberts & Janovy Jr., 2000). Trematode metabolism relies on the breakdown of carbohydrates (primarily glucose and glycogen). Unlike other organisms that utilize most of the energy potential of a carbohydrate, trematodes incompletely catabolize their energy rich resources. Scientists have no definite explanation as to why they do this, but hypothesize it might be due to the relatively inexhaustible supply of nutrients the host provides to the parasite. The miracidia and cercariae do not feed, and thus can only last a few hours (12-24 h) without a host (Roberts & Janovy Jr., 2000). There is likely very strong selective pressure for effective host finding mechanisms (discussed more in behavior section).

Behavior

There is little knowledge concerning the perception of *E. trivolvis* in particular, but there are studies of host perception and host-finding behavior in other trematodes (Thiemann & Wassersug, 2000). Trematodes have several mechanisms of finding a host, which include the use of chemo-orientation, shadow stimuli, touch, and water turbulence. The cues that allow trematodes to distinguish between host and non-host in chemo-orientation are driven by the recognition of molecules given off by the host (Fried & Graczyk, 1997). Since the free swimming stages, miracidia and cercariae, of trematodes are exposed to the environment, they must find a host quickly. This suggests that trematodes engage in some sort of host-finding behavior (Fried & Graczyk, 1997). Miracidia will follow snail host excretory-secretory (ES) chemicals to find them. Products of ES are macromolecules, which can stimulate trematode activity into finding their host. *E. trivolvis* has been seen to respond to products such as, acetic acid, sulfuric acid, and hydrochloric acid, by gathering near the source. Specific chemicals vary depending on

the species. Although more research is being conducted on miracidial host-finding behavior, scientists are still unsure as to what actually causes miracidia to attach to gastropod hosts (Haas et al., 1995). Cercariae also exhibit chemo-orientation host-finding behavior. However, they respond to smaller molecules, like amino acids, instead. Although sharing the same genes, miracidia and cercariae utilize different chemicals to achieve the same objective, finding and penetrating the host (Haberl et al., 2000).

Parasites can affect the behavior of their hosts. Levri (1998) showed that snails infected with *Microphallus*, a genus of trematode, expose themselves to predation risk more often than those that were uninfected. This exposure would allow the internal parasite to pass from one host to another, by increasing the probability that its host would be consumed. On the other hand, this sometimes does not result in the most advantageous outcome, especially if its current host is consumed by a predator that is not the next host in its life cycle (Levri, 1998).

Ecosystem Roles

While largely unexplored empirically, parasites like *E. trivolvis* may play important roles in regulating the abundance and distribution of particular host species. In particular, parasitism may interact with other stressors like predation or pollution (Thiemann & Wassersug, 2000). Tadpoles can differentiate between infected and uninfected snails and move away from infected snails. This helps avoid being penetrated by cercariae that seek them out as a host. However, with the addition of a predator, tadpoles decreased their movement, and exhibited an increase in trematode infection. This has important implications when assessing increased fish (predators) introductions into ponds and lakes where snails and trematodes are present. These introductions may result in an elevation of trematode infections and thus a decrease of native amphibian populations. Pollution can also play a role in parasite-host interactions (Budischak et al. 2008). Exposure to pesticides damages the immune system, leading to an increase in susceptibility to trematodes infections. Eutrophication caused by pollution stimulates snail growth and parasite prevalence within the snail. However, these snails may lack healthy immune responses to prevent parasite infection and thus become a host. This field of parasitology is important when considering global climate change, which may cause an increase in parasite numbers (stimulated by longer reproductive seasons, ideal warmer conditions, and larger habitat ranges) because of a warmer climate (Budischak et al. 2008).

E. trivolvis is not of conservation concern, but trematodes that infect humans have been studied extensively (e.g., schistosomes). However, relatively little is known about trematode effects on amphibians (Fried et al. 1997) or other wildlife hosts. Recently, trematode infections have been linked to declines in some amphibian populations. When some trematodes, like *Ribeiroia ondatrae*, encyst inside an amphibian, they can cause kidney damage and limb deformities (Johnson et al., 1999). Studies conducted by Holland et al. (2006) have shown that frog tadpoles have an elevated risk of mortality, as well as increased edema rates, if exposed to cercariae of Echinostomes. Tadpoles exposed early in their development were more likely to have higher percentages of mortality, than those further along in development (Holland et al., 2006). In addition, pesticides may lower immunity and raise susceptibility to trematode

infections (Rohr et al., 2008). Amphibians, then, may be facing numerous interactive threats (Rohr et al., 2008). More studies need to be conducted to provide data for support.

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