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# Alteration in behavior of intermediate hosts caused by larval stages of some digenetic trematodes

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## I. Abstract

Metacercariai of digenetic trematodes can induced behavioral changes in their intermediate hosts. these effects make infected invertebrate more attractive to the definitive host which fed on it ,this facilitate the transmission of the parasite to the definitive host to complete the life cycle . The current study highlighted the effect of the *Cyclocoelum mutabile* metacercariai on the response of the *Lymnaea auricularia* snails to phototaxis and geotaxis . According to the result of the present study, both infected and un-infected snails are attracted to light, The presence of each of them is concentrated in the two neighboring regions to the source of light ,While a significant differences ($P \leq 0.05$) noticed between the response of infected snails and uninfected snails to earth gravity.

**Key words:** Trematodes, snails**,** Behavioral changes .

## II. Introduction

The effects result from parasitic infections vary in intermediate and definitive hosts , parasites may lead to illness or death of their hosts due to their direct pathological effects on vital tissues and organs or it may induced physiological and behavioral changes in these hosts(Nicolas *et al*. , 1996 ; Barber and Wright ,2006).

The larval stages of parasitic worms have adifferent effects on their invertebrates intermediate hosts represented by direct pathological changes and weakness in resistance to predation as well as other changes that lead to the synchronization of the presence of the intermediate host at the same time or in

the same place where the definitive host is located, in addition to some of the effects that make the infected invertebrates more attractive to the definitive host which fed on them to facilitate the exposure of definitive host to parasites and complete the life cycle ( McNeil and Díaz , 1995 ;Levri,1999). Some of these behavioral changes occur due to the effects of the parasites on the responsiveness of their hosts to some of the environmental factors such as light and gravity ( Levri *et al*. , 2007).

The identification of the effect of the *Cyclocoelum mutabile* metacercariai on the response of their intermediate host to phototaxis and geotaxis is the objective of this study.

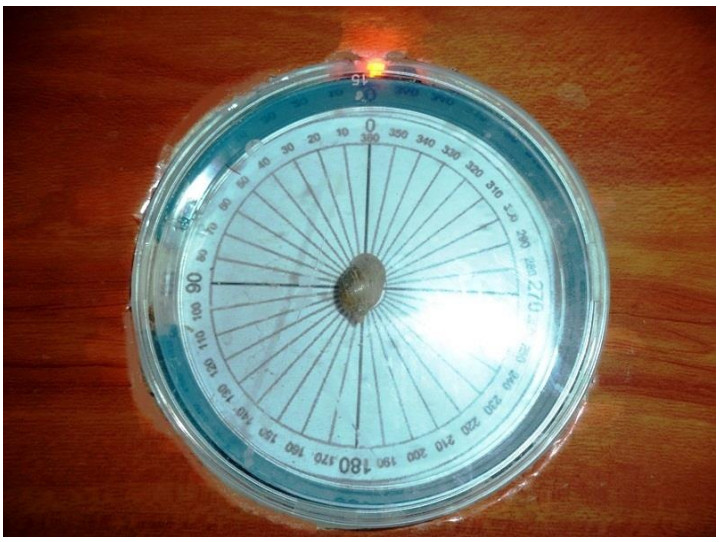
**Methodology**

Samples of *Lymnaea auricularia* snails were collected from the Al –Hamar marsh in Thi Qar province manually, They were placed in plastic containers with a quantity of river water and some aquatic plants and were brought to the laboratory , classification of snails confirmed according to Clarke(1981) and Burch(1989). The snails maintained in an aquarium in the laboratory to obtain a lab population according to McKindsey (1993) .

Seven of *Fulica atra* birds purchased from the vegetables market in Al –Nassiriyah in February 2017 were used as a source of *C. mutabile* trematodes. Birds were killed and trematodes were isolated from the body cavities of infected birds. Trematodes classified according to Kanev *et al*. (2002), Each trematode was placed in an watch glass containing 2 ml of chlorine-free water. It was killed by pricking it with a needle under the head area and opened with a sharp blade to extract the eggs that were left to hatch at a room temperature for obtaining the meracidiai.

A group of laboratory population of *L. auricularia* which ranged in size from 5-7 mm was selected for using in experimental infection which was conducted according to McKindsey and McLaughlin (1995) , each one of these snails placed in well of twelve well tissue culture plates containing 5 ml of chlorine-free water , then each one of these snails exposed to infection by adding three meracidiai to each well,the tissue culture plates incubated at room temperature for 24 hours ,then the snails transferred to 500 ml plastic containers were fed and kept under the same laboratory conditions at 25 ° C . A group of laboratory generations of *Lymnaea auricularia* from the same size was placed in the same conditions (unless exposure to infection) for using it as control group.

**The effect of infection on response of snails to phototaxis experiment** Thirty of *Lymnaea auricularia* snails after forty days of exposure to infection were selected for this experiment . the experiment was done according to Levri and Fisher (2000) with some modification, a paper circle divided to equally tagged lines ( from 0 to 360 ) was used, a petry dish with chlorine-free water at room temperature set on the circle , A wooden box set over this dish for blocking light. A source of fiber optic light used to produce beam of light which shine in the box from the 0 line direction by a small hole in the box( figure, 1), each of exposed snails placed separately in the center of the dish and left for ten minutes .then ,the box removed to record the location of the snail. At the end of the experiment, all these snails necropsied to confirm the infection, the experiment returned by using thirty of snails from control group .



**Figure(1) The circle which used in phototaxis experiment**

**The effect of infection on response of snails to Geotaxis experiment**

Thirty of *Lymnaea auricularia* snails after forty days of exposure to infection were selected for this experiment . The experiment was conducted in the morning with natural lighting according to Levri and Fisher (2000) with some modification, a graduated glass tube (200mm long and 5mm wide) was used , after filling the tube with chlorine-free water at room temperature , The exposured snails placed individually at the center mark of the tube and when the snail attached itself to the center mark of the tube, the tube placed vertically in a 25x25x25 cm glass basin filled with chlorine-free water at room

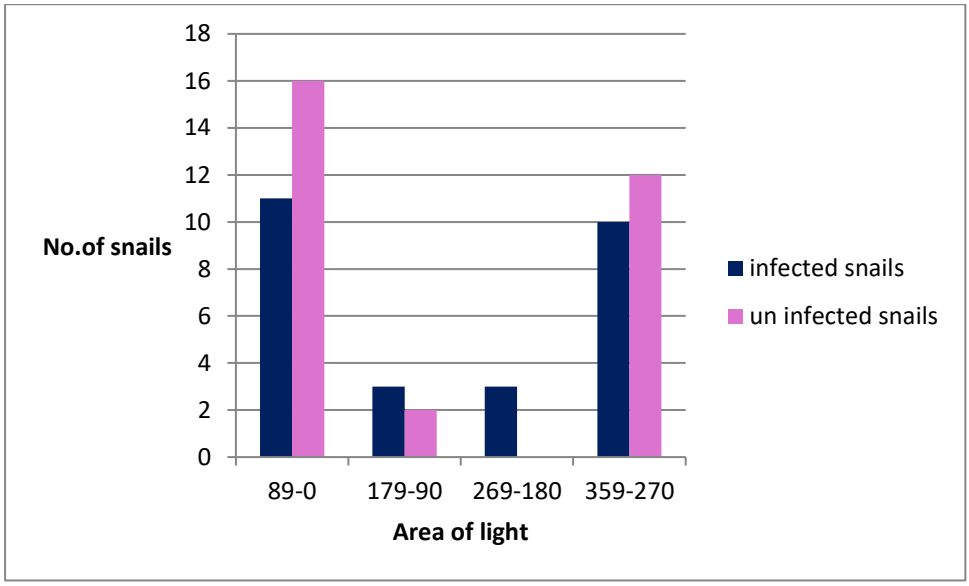
temperature 25 ° C and leave it for 10 minutes, the tube pulled and the direction and distance of snails movement recorded. The experiment returned by using thirty of snails from control group. The experiment returned by using thirty of exposured snails and then by using thirty snails from control group at dark conditions(a glass basin covered tightly by using a dark cover). At the end of the experiment, all these snails necropsied to confirm the infection.

The data were statistically analyzed using t-test for the comparison between the two groups ( SPSS version 18), to determine if there was a significant differences in the response of infected snails and the response of un-infected snails to phototaxis, as well as the significant differences between the response of infected snails and the response of uninfected snails to the geotaxis.

## III. Results

**The effect of infection on response of snails to phototaxis experiment** The dissecting of the snails used in this experiment, which was previously exposed to infection, showed that 27 of them were infected with metacercariai .

statistical analysis referred to a significant increase($P \leq 0.05$) in the number of both of infected and uninfected snails which moved towards the source of light as a comparative with both infected and uninfected snails which moved away from the light source ,while the number of infected snails which moved to the source of light did not differ significantly ($P \leq 0.05$) from the number of uninfected snails which moved to the source of light, that's mean, both infected and un-infected snails are attracted to light, The presence of each of them is concentrated in the two neighboring regions to the source of light(11 snails in the area between 0 line to 89 line and 10 snails the area between 270 line – 359 line) ( figure ,2).




**Figure( 2) Distribution of of infected and uninfected snails around areas of light source**

**The effect of infection on response of snails to Geotaxis experiment**

The dissecting of the snails used in this experiment, which was previously exposed to infection, showed that 28 and 27 of them were infected with metacercariai in light and dark conditions respectively. The response of infected snails to geotaxis was negatively affected in both light and dark conditions compared to uninfected snails ,21out of 28 and20out of 27infected snails didn't react with geotaxis in light and dark conditions respectively , by using statistical analysis , a significant differences ($P \leq 0.05$) noticed between the response of infected snails and uninfected snails to earth gravity (down orientation) (Figure ,3).

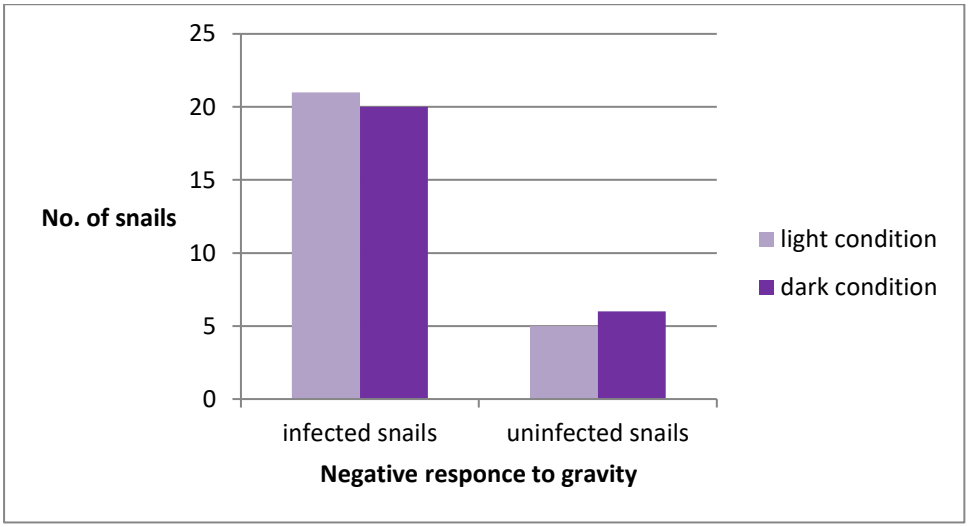



**Figure (3) Number of infected and uninfected snail Respond negatively to geotaxis in light and dark conditions**

## IV. Discussion

Parasites often cause some behavioral changes in their intermediate host to make them more susceptible to discovery and predation by final host (Kaldonski *et al*., 2007 ) , In particular the effect on the response of light and gravity (Bauer *et al*.2005). Light is one of the most important factors that some mollusks adopt in determining the direction of their movement (Warburton, 1973; Van Duivenboden , 1982) and according to the results of this study , both infected and un infected *L.auricularia* snails with *C. mutabile* metacercariai were attracted to light and moved toward it , This means that the infection did not have an effect on this characteristic Which is a natural recipe in these snails. The conclusion of Wesołowska and Wesołowski (2014) on the effect of parasitic infection on the response of intermediate host to phototaxis was related to the land snails *lnus glutAinosa* which acts as an intermediate host for *the Leucochloridium* trematodes , these snails live among the terrestrial plants and are not attracted to the light naturally making their discovery difficult by birds which acting as final hosts to those trematodes, the larvae of these worms make changes in the behavior of these snails, so that they are attracted to light and move towards it and go up to the top of the plants to facilitate their predation by birds.

The results of this study revealed that metacercariai of *C. mutabile* have a negative effect on *L. auricularia* snail's response to gravity, This behavioral change may be beneficial to the parasite, so that the infected snails be easy to catch by the final hosts of these trematodes, The results of this study correspond with the results of the field study of Levri (1999) Which it conducted on *Potamopyrgus antipodarum* snails,the intermediate hosts of *Microphallus* trematodes , he recorded that uninfected snails react positively with geotaxis ,hiding in the lowlands and under the rocks to avoid predation while the interaction of infected snails with gravity is not found So they exist more frequent on the tops of rocks to facilitate their discovery by birds. In the laboratory study conducted on the same snails by Levri and Fisher(2000), The researchers found that the infected snails with *Microphallus* lose their ability to interact with the earth's gravity. These behavioral changes due to parasitic infection may return or be linked with ability of parasites to alter the chemical communication of their hosts Klein( 20

## V. Refernces

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