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# METACERCARIAL INFECTION IN THE MUSCLES OF TILAPIA NILOTICA AND CLARIAS LAZERA AND THE EFFECT OF SOME PHYSICAL AND CHEMICAL FACTORS ON THE VIABILITY OF ENCYSTED METACERCARIAE.

(With 4 Figures and 3 Tables)

By

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## إصابة عضلات أسماك البلطي والقرموط باليرقات المتحوصلة لديدان التريماطودا وتأثير بعض العوامل الطبيعية والكيميائية على حيوية هذه اليرقات

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بفحص عضلات أسماك البلطي والقرموط لوجود الأطوار اليرقية المتحوصلة لديدان التريماطودا تبين وجود نوعين من هذه اليرقات في سمك القرموط ونوع واحد في سمك البلطي. وبعدها حيوانات التجارب بهذه اليرقات تم الحصول على الأطوار البالغة لها وكانت لديدان السينوديبيلوستوم ازيمي ( من القراميط ) والبروهيمستوم فيفاكس ( من القرامط والبلطي والبلطي ) وتم دراسة التأثيرات المختلفة مثل التبريد والتجميد والشى وحمض السيتريك على حيوية هذه اليرقات المتحوصلة في العضلات لمعرفة أفضل الطرق لقتلها حتى لاتضر بصحة الانسان.

\* : Part of M.V.Sc. Thesis

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## SUMMARY

Macroscopic encysted metacercariae in muscles of *Clarias lazera* were proved to be the larval stage of *Cynodiplostomum azimi*, while the microscopic ones obtained from both *Clarias lazera* and *Tilapia nilotica* were found to be those of *Prohemistomum vivax*. Both parasites were recorded in man. Metacercariae in *C. lazera* were destroyed by chilling at (4°C) after 72 hours and by freezing at (-10°C) after 24 hours. Metacercariae in *Tilapia nilotica* were killed after 48 hours chilling and 24 hours freezing. Grilling killed all metacercariae in *Tilapia nilotica* within 15 minutes. 10% citric acid killed metacercariae in *T. nilotica* within 24 hours but those in *C. lazera* were killed within 48 hours, 20% citric acid killed metacercariae in both fishes within 24 hours.

## INTRODUCTION

A special references will be pointed out about the dangers of fish as a source of transmitting parasites to man and fish-eating mammals and the diseases caused by such parasites. NASR (1941) recorded *Prohemistomum vivax* in man. HAMED and ELIAS (1970) reported that when man eats insufficiently cooked fish, the metacercariae reach the small intestine and mature there causing abdominal discomfort. EL-NAFFAR (1970) recorded encysted metacercariae of *Prohemistomum vivax* in the muscles of *Tilapia* spp. FAHMY et al. (1976) studied the encysted metacercariae in *Tilapia* and *Clarias lazera* in Assiut province. FAHMY et al. (1980) noticed the effect of freezing on metacercariae of *Prohemistomum vivax* and *Haplorchis pumilio*. MAHMOUD et al. (1988) studied the effect of diluted acids (commercial vinegar 6% and lemon juice) on metacercariae of *Prohemistomum vivax* in Sohag province.

The present study was conducted to identify the different types of encysted metacercariae in the muscles of the most common species of Fishes in Assiut City and the most reliable methods for killing these encysted metacercariae.

## MATERIAL and methods

Fifty *Clarias Lazera* and fifty *Tilapia nilotica* Fishes were collected from Fish markets at Assiut City. Their Muscles were



examined grossly and Microscopically for the presence of metacercariae. The metacercariae which found were fed to Laboratory mice and adults were collected from their small intestine within one week after infection. Both metacercariae and adults were then Fixed in 70% alcohol, stained with acetic alum carmin and mounted in Canada Balsam. They were identified according to the Keys presented by Yamaguti (1958). Viable Metacercaria were recognized by their movements inside their cysts and by experimental infection to mice and detection of adult flukes from the small intestine of mice sacrificed seven days post-infection. Pieces of fish muscles which contain viable metacercariae were exposed to chilling (4 C), Freezing (-10 C) for 24, 48, 73 Hours, grilling (For the Muscles of *T. Nilotica*) and concentration of 10% and 20% citric acid for 24 and 48 Hours. Viability of the treated metacercariae was studied by the previously mentioned methods.

## RESULTS

Two types of encysted metacercariae were detected in the muscles of *Clarias Lazera*. The first type Appeared grossly as a small white nodules (Macroscopic type). The other type was detected only by microscopic examination (Microscopic Type). Fig. (1, 2). Microscopic type of encysted metacercariae revealed adult flukes of *Cynodiplostomum azimi*, NAZMI (1933) Fig. (3), while the microscopic type revealed the adult worm of *Prohemistomum vivax*, SONSINO (1892) FIG. (4). IN CASE OF *Tilapia nilotica* one type of encysted metacercariae was detected which yielded in mice the adult of *Prohemistomum vivax*.

Effect of chilling and freezing on the viability of encysted metacercariae in *Clarias lazera* and *Tilapia nilotica* is shown in tables 1, 2.

Grilling of pieces of muscles of *Tilapia nilotica* for fifteen minutes was enough to kill the encysted metacercariae present in them.

Effect of 10% and 20% citric acid solutions on the viability of encysted metacercariae in *Clarias lazera* and *Tilapia nilotica* is shown in table 3.

## DISCUSSION

The present study indicates that both *Tilapia nilotica* and *Clarias lazera* fishes act as a second intermediate host for *Prohemistomum vivax*. This agree with the finding obtained by EL-NAFFAR (1970), who proved that encysted metacercariae found



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in *Tilapia nilotica* were the larval stage of *Prohemistomum vivax*. Also, FAHMY *et al.* (1976) and OTIFY (1989) proved that *Tilapia nilotica*, *Tilapia zilli* and *Clarias lazera* act as second intermediate hosts for *Prohemistomum vivax*.

In the present study the macroscopic type of encysted metacercariae produced the adult flukes of *Cynodiplostomum azimi*, NAZMI (1933). This agree with KHALIFA *et al.* (in press). They described two types of metacercariae in muscles of *Clarias lazera* and obtained the adult flukes of them, *Cynodiplostomum azimi* and *C. deboisi*. EL-SHERBINY (1987) identified diplostomatid metacercaria from the muscles of *C. lazera*, he Regarded the effect of chilling (4°C) and freezing on the viability of encysted metacercariae, the results obtained in the present study were similar to those recorded by GHIHINE (1972), ROBERT (1978), BROWN & NEVA (1983). They noticed that deep freezing of fish muscles at -10°C for 24 hours was sufficient to destroy the encysted metacercariae.

Regarding the effect of grilling on the viability of metacercariae, HAMED and ELIAS (1970) found two viable metacercariae in the muscles of Mulletts after grilling. This result in not in agreement with the present study in which all metacercariae were killed after 15 minutes of continuous grilling. This may be attributed to the difference in the time used for grilling or the type and thickness of fish used.

As regarde the effect of different dilutions of citric acid (10%, 20%) on the viability of encysted metacercariae, in case of *Tilapia nilotica* the present results agree with that obtained by MAHMOUD *et al.* (1988), they reported that the dilution of citric acid (100%, 50%, 25% and 5%) were lethal to all metacercariae in muscles of *Shilbe mystis* exposed for 24 hours. The present data were, however, different in case of metacercariae in *Clarias lazera* which were destroyed within 48 hours in 10% citric acid. This was attributed to the type of encysted metacercariae and their resistance to the effect of acid.

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- Fig. 4: *Prohemistomum vivax*, adult worm, from the intestine of experimentally infected mice.

Table (1): Effect of chilling and freezing on the viability of encysted metacercariae in muscles of *Clarias lazera*.

Time/h	Chilling		Freezing	
	Temperature	Viability	Temperature	Viability
0.00	25 °C	viable	25 °C	viable
24	4 °C	viable	-10 °C	Dead
48	4 °C	viable	-10 °C	Dead
72	4 °C	Dead	-10 °C	Dead

Table (2): Effect of chilling and freezing on the viability of encysted metacercariae in muscles of *Tilapia nilotica*.

Time/h	Chilling		Freezing	
	Temperature	Viability	Temperature	Viability
0.00	25 °C	viable	-10 °C	viable
24	4 °C	viable	-10 °C	Dead
48	4 °C	Dead	-10 °C	Dead
72	4 °C	Dead	-10 °C	Dead

Table (3): Effect of some different dilution of citric acid on the viability of encysted metacercariae in *Clarias lazera* and *Tilapia nilotica* stored at room temperature.

Time/h	<u>Clarias lazera</u>		<u>Tilapia nilotica</u>	
	Citric acid			
	10%	20%	10%	20%
0.00	Viable	Viable	Viable	Viable
24	viable	Dead	Dead	Dead
48	Dead	Dead	Dead	Dead



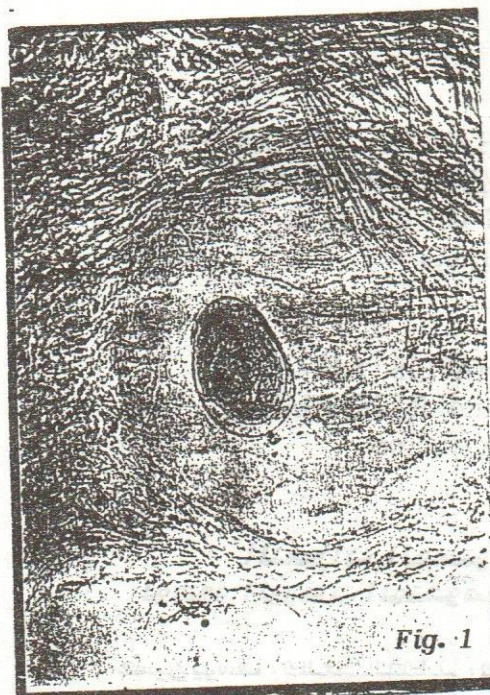


Fig. 1



Fig. 2

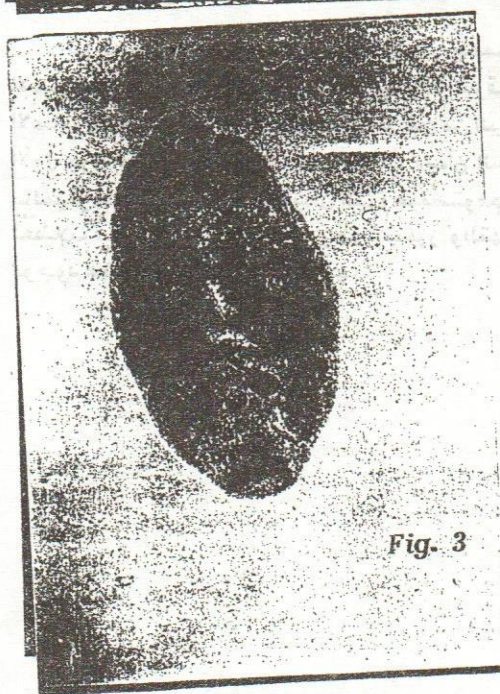


Fig. 3



Fig. 4