Fish Farms as a source for parasites transport: Parasitological and developmental studies of *Prohemistomum vivax* with the ameliorating role of *Moringa oleifera* in the treatment

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Abstract: Fishes are good source of quality protein, but various diseases including parasitic infections pose a threat to fish culture. Catfish, *Clarias gariepinus* is one of most famous fishes in fish farms that infected with *Prohemistomum vivax* metaceracriae. *Prohemistomum vivax* was rarely recorded to infect human and may cause death. This work aimed to study the development to adult worms in vivo, its maturation, recovery rate, time of appearance of the first egg in the stool of infected rats, the percentage of recovered worms and distribution of the parasite within the intestine of the host. In addition to the ameliorating role of *Moringa oleifera* in the treatment of experimental infected rats with metacercaria of *Prohemistomum vivax*. The experiments were performed on 70 male albino rats weighing 100 ±10g and of 7-8 week's age. Encysted metaceracriae were isolated, counted and force feeidng infection was made by a blunt forceps. Rats were infected with 500±50 metaceracriae and only three infected rats were scarified at different internals (1hr, 3hr, 6hr, 12hr, 1 day, 2 day, 3 day and 1 week). After one week of infection; rats were treated with moringa for five days. Our results shows that the encysted metacercariae of *Prohemistomum vivax* were scattered in between the muscle fibers of the head, trunk and caudal regions of naturally infected catfish. Three days were enough to adult worm development and the 1st eggs were detected in the stool after 3 days. The recovery rate was significantly decreased with the increase of treatment time. Duodenal sections in infected rats with *Prohemistomum vivax* showed villi were marked epithelial compression and erosion, atrophy, hemorrhage, inflammatory cells infiltration of mucosal stroma, cytoplasmic vacuoltions, congested blood vessels, goblet cell depletion and crypt hyperplasia. No adult worms were appeared in the rat stool before treatment whoever after treatment with moringa, the adult worms were detected in 2nd day of treatment and completely disappeared at the 4th days of treatment.

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1. Introduction

Fishes are good source of quality protein, but various diseases including parasitic infections pose a threat to fish culture (Yooyen et al., 2006). In addition to the economic loss to farmers, many of the parasites, particularly trematodes, are also of zoonotic importance. Eating raw or, improperly cooked or processed fish is the main source of these infections to human that has been reported from various geographical regions (Park et al., 2009), causing laryngitis. Digenetic trematodes and their metacercariae take a great interest in most countries especially for the human care against the transmissible diseases (Taher, 2009; Hassan et al., 2012). They were considered as one of the most common parasites infecting fish causing low weight gain, high mortality, immarketability and some of these parasites may have zoonotic importance (Hernandez *et al*., 1998**;** Taher, 2009).

The World Health Organization (WHO) has estimated that the number of people currently infected with fish-borne trematodes exceeds 18 million, and many more are at risk (WHO, 1995). In great proportion parasites are disseminated and introduced to new localities for the movement of infected host. However, in KSA the anthropogenic interest over any scientific opinion has been the main factor for the introduction and establishment of helminth species in new localities (Scholz *et al*., 1999; Salgado-Maldonado and Pineda-López, 2003, Raef et al., 1999, 2003).

Catfish, *Clarias gariepinus* is one of most famous fishes in fish farms. *Prohemistomum vivax* was rarely recorded to infect human and may cause death (Nasr, 1941; Williams and Jones, 1976). Fahmy et al. (1976*)*; Rifaat et al. (1980) and Shalaby (1982, 1993) identified *P. vivax* from puppies and kittens fed on metacercariae infecting fishes. Shalaby (1985) detected *P. vivax* in intestine of dogs after experimental infection with metacercariae in catfish. Tantawy (1993) identified *P. vivax* from experimentally infected kittens, rats and pigeons with metacercariae infecting fresh water fishes. Raef (1994) obtained *P. vivax* after experimental infection of puppies, chickens, ducks and albino rats with metacercariae infecting marine fishes. Saba (2004)

obtained *P. vivax* after orally administrating the metacercariae in fresh water fishes to puppies, chickens and ducklings.

People have long known that botanical medicine provided a complete, safe system of healing and prevention of diseases. *Moringa oleifera*, is one of the best known and most distributed species of Moringacae family. Moringa is an important tropical crop that is used as human food, medicine and in oil production. It is a small sized tree, which is native to south Asia, Africa and Arabia (Anwer et al., 2007). It is commonly known as drumstick tree or horse radish tree. Medicinally, various parts of Moringa are generally known for their multiple pharmacological effects including their antitumor (Bharali et al., 2003), antihyperglycemic (Anwer et al., 2007) and anti-inflammatory (Mahajan and Mehta, 2008) effects. Furthermore, the extract of Moringa has been shown to have potent antioxidant action in vivo (Kumar and Pari, 2003). Metcercarial infections are the most common parasitic infection of the fish. Despite the importance of metacercaria in the life cycle of trematodes. Hence, the objective of this work is to study the development to adult worms of *Prohemistomum vivax* in vivo, its maturation, recovery rate, time of appearance of the first egg in the stool of infected rats, the percentage of recovered worms and distribution of the parasite within the intestine of the host. Also, the histological changes in rat intestine after infection. In addition to the ameliorating role of *Moringa oleifera* in the treatment of experimental infected rats with metacercaria of *Prohemistomum vivax.*

2. Material and Methods

Metacercariae of *Prohemistomum vivax* was isolated from the skeletal muscles of naturally infected catfish, *Clarias gariepinus* that were collected from the River Nile in Egypt. Encysted metacercariae were used for the infection of the albino rats. The experiments were performed on 70 male albino rats (*Rattus norvigicus*) weighing 100 ±10g and of 7-8 week's age. They were obtained from our University farms. The rats were kept in the laboratory for one week before the experimental work and maintained on a standard rodent diet and water available *ad libitum*. The temperature in the animal room was maintained at 23±2^{O}C with a relative humidity of 55±5%. Light was on a 12:12 hr light -dark cycle. The experimental protocol was approved by Local Ethics Committee and Animals Research.

Isolation of metaceracriae from infected *clarias lazera*: The catfish were decapitated, skinned and the muscles were examined for the presence of *Prohemistomum vivax* metaceracriae. This was made by squeezing a piece of muscle between two glass slides and the examination under a dissecting microscope. Encysted metaceracriae were mechanically removed, isolated, counted with the aid of a dissecting microscope they were then withdrawn by a Pasteur pipette and kept in 0.75% saline solution for the infection of rats (Khalil 1987).

Experimental infection of laboratory animals: Encysted *Prohemistomum vivax* metaceracriae were isolated, counted and force feeidng infection was made by a blunt forceps (Khalil 1987). Rats were infected with 500±50 metaceracriae and only three infected rats were scarified at different internals (1hr, 3hr, 6hr, 12hr, 1 day, 2 day, 3 day and 1 week) and adults *Prohemistomum vivax* were recovered from their intestine. After one week of infection; rats were treated with moringa for five days.

Histopathological investigations: Small pieces from control and infected catfish with metacercaria and also immediately after decapitation rats were dissected and pieces of small intestine were removed and fixed in 10 % neutral buffered formalin. Fixed muscles and small intestine specimens were dehydrated in an ascending series of alcohol, cleared in two changes of xylene and embedded in molten paraffin (mp. 50–58°C). Sections of 5 microns thickness were cut using rotary microtome and mounted on clean slides. Sections were stained with Ehrlich's haematoxylin and counterstained with eosin as a routine method after Bancroft and Stevens (1990).

3. Results and Discussion

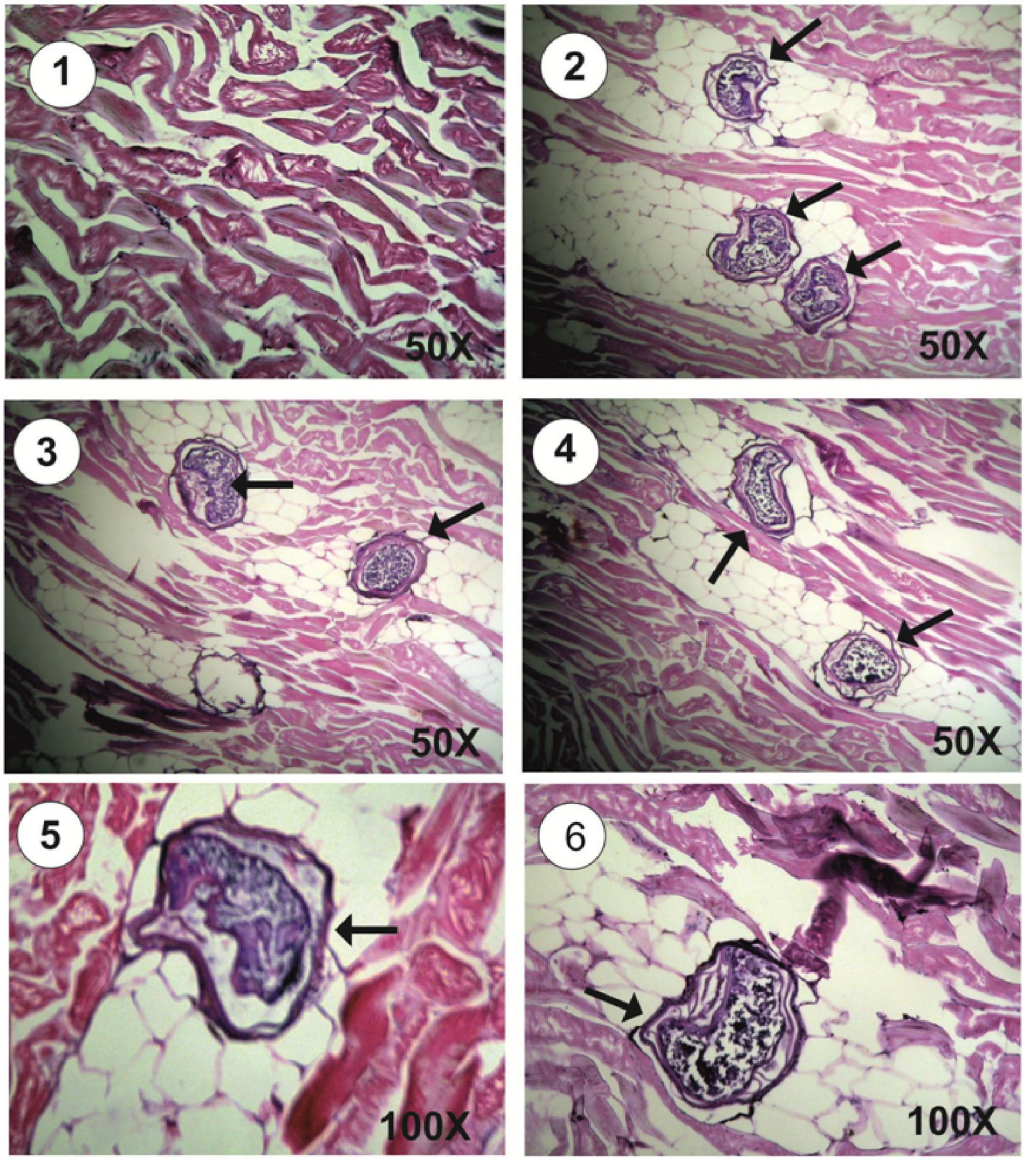
The encysted metacercariae of *Prohemistomum vivax* were scattered in between the muscle fibers of the head, trunk and caudal regions of naturally infected catfish, *Clarias gariepinus*. These are measuring 290-320 by 300-340 microns in size (Figures 2-6). They are subspherical, double walled, outer thick and inner hyaline and separated from the metacercaria by a potential space containing fluid in which the metacercaria was moving, vacuoles and adipose tissues were present in this space (Figures 2-6) when compared with normal control muscles (Figure 1). The encysted metaceracriae were surrounded by a thick layer and it was usually folded inside its cyst wall (Figures 4-6).

The development of adult worms from the encysted metacercaria that isolated from the skeletal muscles of naturally infected catfish, *Clarias gariepinus* was presented in Figure (7-27). After 1 hour of infection, Most of metacercaria were excysted (64%) and the rest were encysted (Figures 7-10), after 3 hours, all infected metacercaria were excysted, the oral sucker and acetabulum were

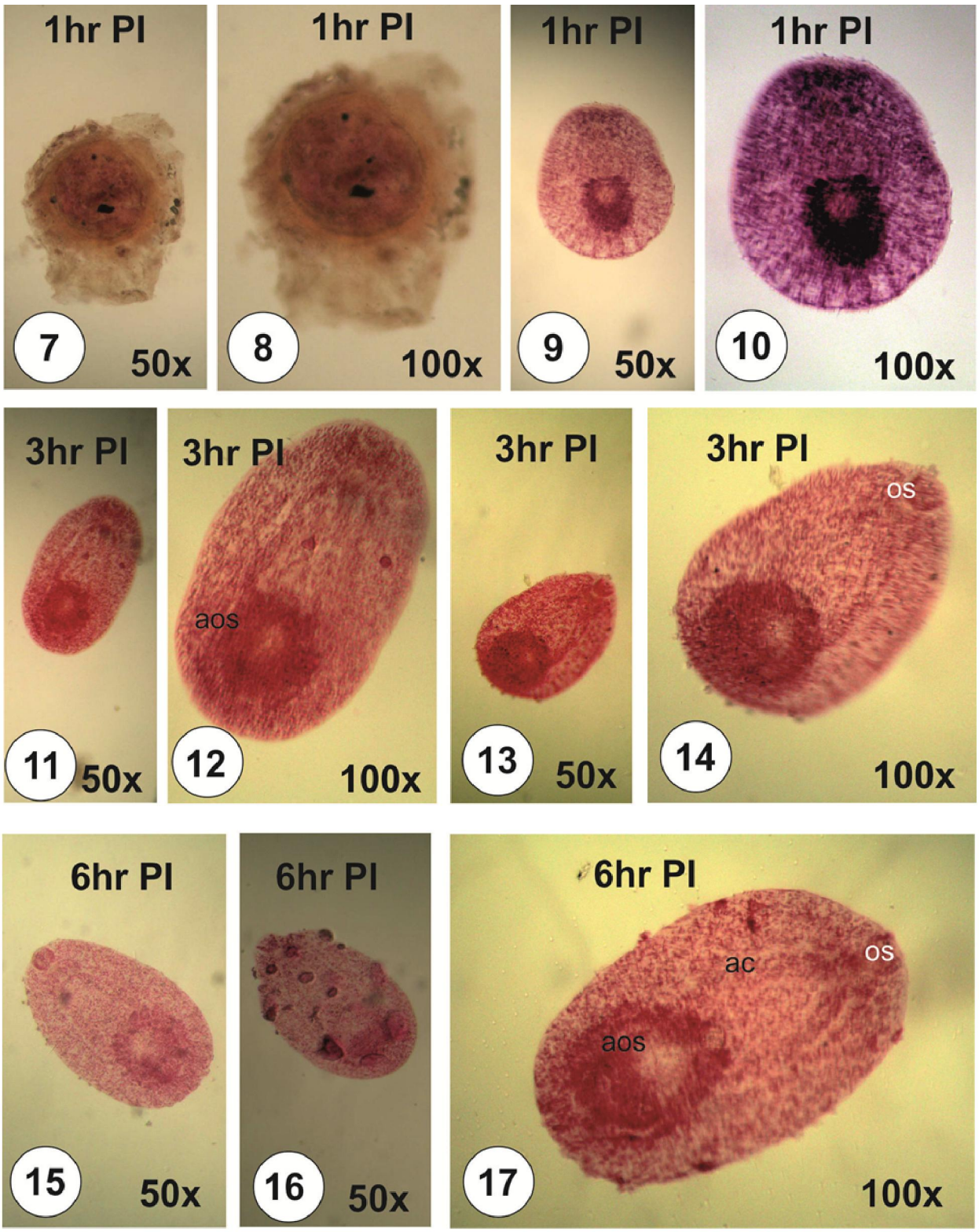
observed (Figures 11&12). After 6, 12 hours and 1 day of infection, only the alimentary canal were detected (Figures 13-19) while after 2 day of infection, the reproductive organs were observed (Figures 20&21). After 3 days of infection, the adult worms were completely well developed with the presence of one or two eggs in uterus (Figures 22-24). After 4 days of infection, the adult worms were completely well developed with the presence of two to four eggs in uterus (Figures 25-27).

Table 1: Recovery rate and changes of eggs and adult worms in rats stool after infection and treatment.

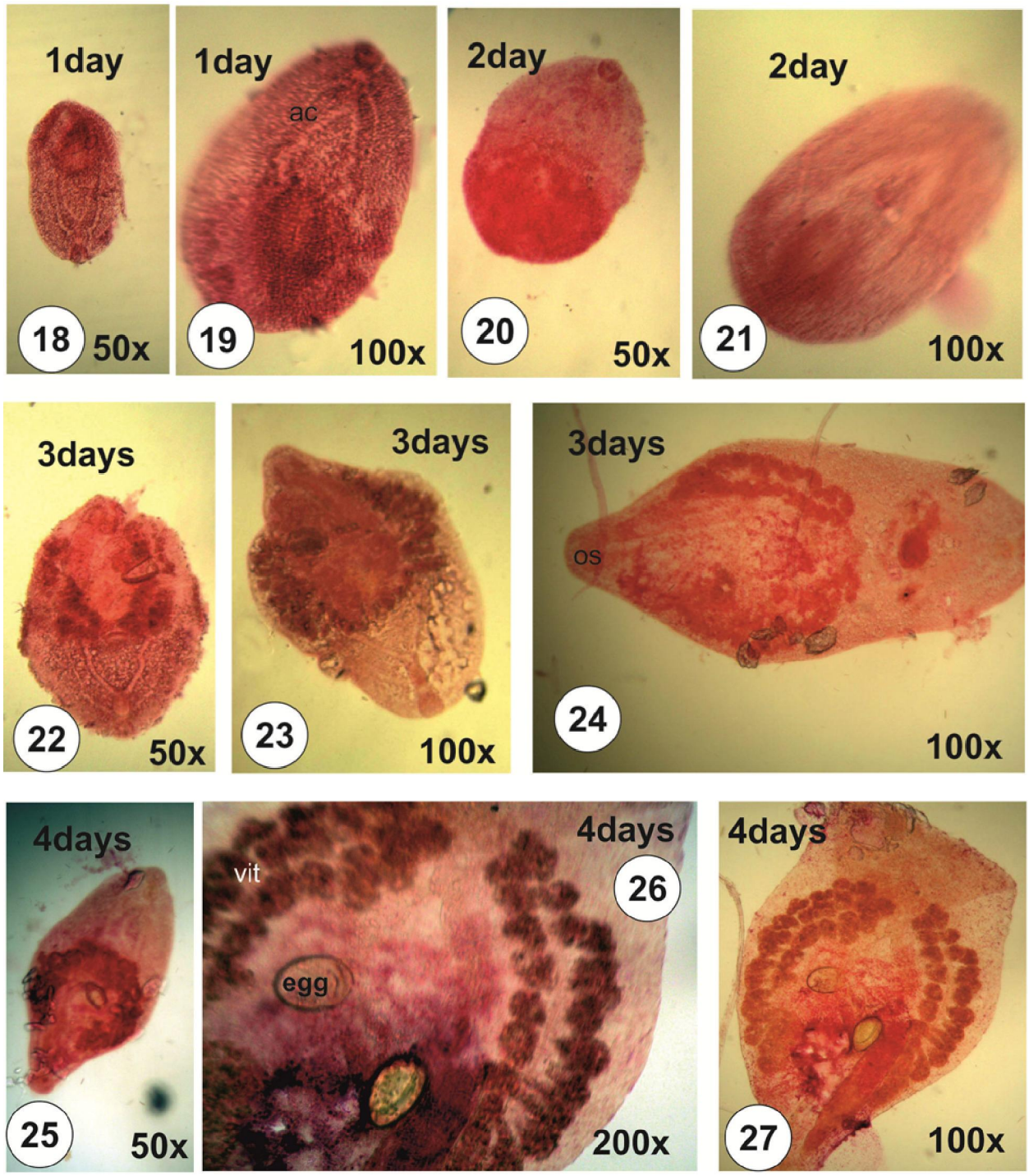
	3rd day of infection	1st day of treatment	2nd day of treatment	3rd day of treatment	5th day of treatment
Recovery rate	76% (758±10)	63% (630±10)	41% (410±10)	29% (285±10)	0% (No worm recovered)
Eggs in stool	Presence	Presence	Presence	Absence	Absence
Adult worms in stool	Absence	Absence	Presence	Presence	Absence



Figures 1-6: Photomicrographs of Hematoxylin-eosin-stained metacercaria in catfish skeletal muscles. **1:** Control skeletal muscles in catfish. 2-5: Infected skeletal muscles in catfish with metacercaria of *Prohemistomum vivax*.



Figures 7-17: Photomicrographs of the development of adult worms from the encysted metacercaria in rat (stained with Hematoxylin-eosin).



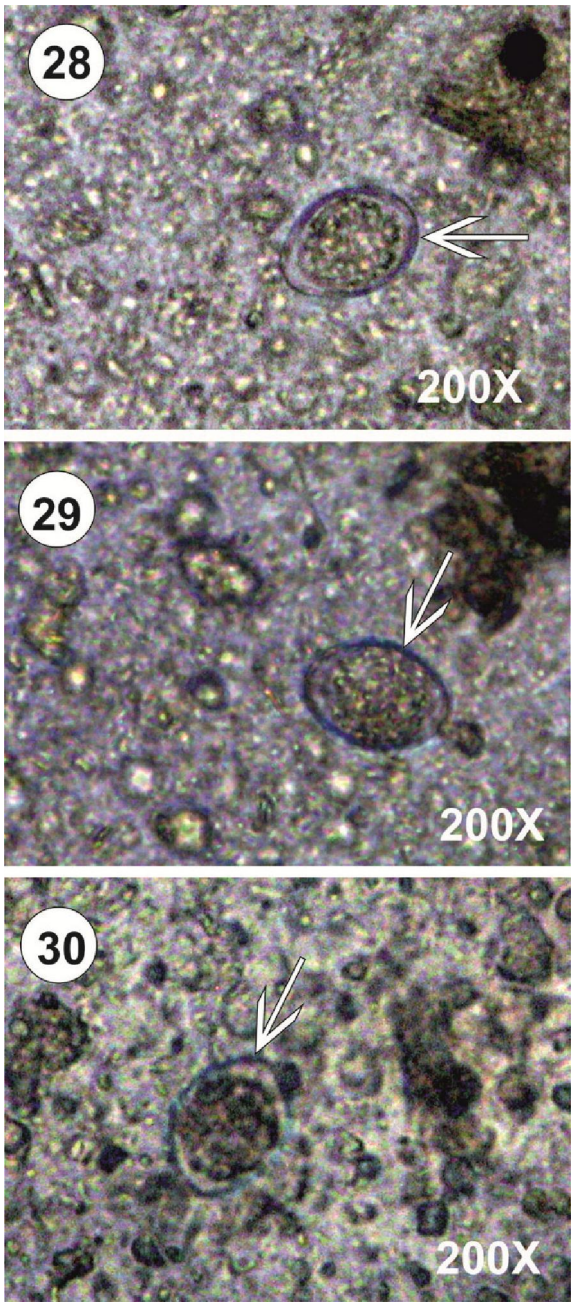
Figures 18-27: : Photomicrographs of the development of adult worms from the encysted metacercaria in rat (stained with Hematoxylin-eosin).

The adult worms of *Prohemistomum vivax* are pyriform, convex dorsally and concave ventrally behind the middle half of the body. The total length of the worm is 1.10-1.51 mm and 0.47-0.69 mm in breadth. The oral sucker is rounded, subterminal and measures 0.05-0.07 mm long by 0.06-0.08 mm wide, while the acetabulum is rounded and measures 0.03-0.045 mm by 0.043 - 0.048 mm. The pharynx is oval in shape and measures 0.042- 0.072 mm long by 0.04-0.08 mm wide. The oesophagus leads to two intestinal caeca ending at a level posterior to the posterior testes. The ovary is small, rounded, lies

posterolateral to the anterior testes and measures 0.11-0.23 mm long by 0.09-0.25 mm wide. The vitellaria are well developed, with two large testes were found and are tandem in position with a narrow intertesticular space. The cirrus pouch is saccular shaped and contains the seminal vesicle and rod shaped cirrus. The eggs within the uterus are few 1-3 in number each is large, oval shaped, yellowish in colour and measure 75 – 90 µm long by 40-65 µm wide (Figures 28-30). The prepatent period is found to be 3-4 days (Figures 22-27).

It was difficult to confirm the identification of the different kinds of microscopic metacercariae when they were encysted in fish or after isolation (**El-Naffar and El-Shahawi, 1986;** Noga, 2000). For this reason the microscopic metacercariae collected from various parts of the catfish, *Clarias gariepinus* were pooled and used or experimental infection of albino rats. The present experimental work showed that only one of adult flukes of digenetic trematodes were obtained after 3 days post infection, where their eggs start to be detected from experimentally infected eats. Examination of stained adult worms showed that they are *Prohemistomum vivax* that was identified before by Azim (1938), Fahmy and Selim (1959), Fahmy *et al*. (1976), El-Naffar *et al*. (1985) and Shalaby (1985). The present study indicates that catfish, *Clarias gariepinus* acts as a second intermediate host for *Prohemistomum vivax* while rats act as a final or definitive host. These results support those investigated by previous workers (Fahmy and Selim 1959, Fahmy *et al*., 1976 and El-Naffar *et al*., 1985). The detected *Prohemistomum vivax* is transmisable to man (Nasr 1941, Khalifa *et al*., 1977; Tadros and El-Mokadem 1983; Bowman et al., 2003; Taher, 2009).

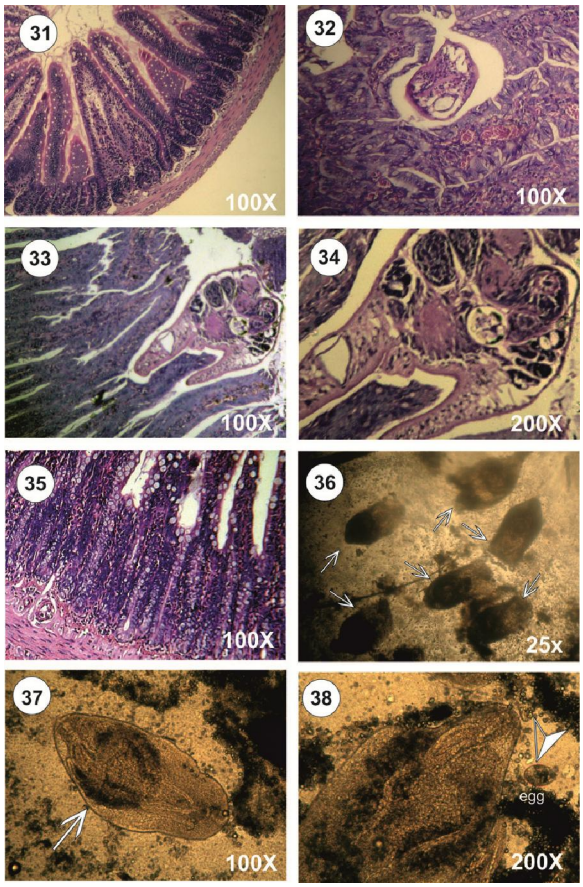
Table (1) shows the percentage of recovered worms of 1000 metacercaria infection was 76% after three days of infection while the recovery rate were significantly decrease after treatment with moringa. The recovery rate was significantly decreased with the increase of treatment time (Table 1). Most of adult worms were detected in the fore intestine especially in duodenum and the rest were detected in proximal ileum. This distribution is completely difference after treatment where most of worms detected in ileum. At the 5th day of treatments the recovery rate was 0%. The 1st eggs were appeared on the 3rd day of infection in the stools (Figures 28-30). After treatment with moringa, the eggs appeared in rat stools for three days and after them no eggs were detected in the stool of treated infected rat (Table 1). No adult worms were appeared in the rat stool before treatment whoever after treatment with moringa, the adult worms were detected in 2nd day of treatment and completely disappeared at the 4th days of treatment.



Figures 28-30: Photomicrographs of the recoverd eggs of infection in the stools.

In agreement with our results; Tantawy (1993) identified *P. genata, Prohemistomum vivax, Procerovum caderoni,* and *M. appendiculatus* from experimentally infected kittens, rats and pigeons with metacercariae infecting fresh water fishes. Raef (1994) obtained *Prohemistomum vivax,* and *M. burmanicus* after experimental infection of puppies, chickens, ducks and albino rats with metacercariae infecting marine fishes. Fayek et al. (1997, 1999) isolated *Microphallus minus* and *Maritrema kitanesis*

after feeding ducklings on metacercariae infecting white shrimp. Saba (2004) obtained *H. heterophyes*, *H. aequalis, P. vivax* and other different types of trematodes after orally administrating the metacercariae in fresh water fishes to puppies, chickens and ducklings. The present results agree with Khalil (1987) who reported that the metacercaria of *P. vivax* required three days to changed to adult worms in final host.



Figures 31-38: Photomicrographs of rat duodenal sections stained with Hematoxylin-eosin. **31:** Control duodenum in rat. **32-34:** Infected duodenum in ratshows the adult worm of *Prohemistomum vivax*. **35:** Treated duodenum with extract of moringa leaf shows no adult worms. 36-38: Removed adult worms of *Prohemistomum vivax* from stool of treated rat extract of moringa leaf.

Section from the duodenum in control rats revealed well preserved mucosal integrity with well arranged finger – like villi and their epithelial linings. The normal features of sub mucosa and other duodenal layers were also seen. Few inflammatory cells were seen to infiltrate into the villous stroma and sub mucosa (Figure 28). The present investigation revealed that dilation and congestion of duodenal blood vessels and stromal lymphocytic infiltration often emerge at considerable distance from the worm praesoma. This agrees with the finding of Thurston *et al.* (1998), who explained such change as a sign associated the inflammatory reactions and infiltration in this layer. Duodenal sections in infected rats *Prohemistomum vivax* showed full maturity of the worm that were seen to entrap the villi by ventral curvature of the fore part of their bodies. In most cases the pathological changes were confined to the mucosal layer (Figures 29&30). Almost all of the villi were marked epithelial compression and erosion, shortened, blunted, thickened, atrophy, hemorrhage, inflammatory cells infiltration of mucosal stroma, cytoplasmic vacuoltions, congested blood vessels, intraepithelial cell infiltration, goblet cell depletion and crypt hyperplasia (Figure 30).

In the current study, we used extract of moringa leaf to treated the infection with *Prohemistomum vivax*. *Moringa oleifera* was claimed to boost immune systems (Olugbemi *et al.,* 2010). The leaves and green fresh pods are used as vegetables by man and are rich in carotene and ascorbic acid with a good profile of amino acids (Makkar and Becker, 1996). They are also used in livestock feed and the twigs are reported to be very palatable to ruminants (Kakengi *et al.,* 2007). *Moringa oleifera* extract was reported to have antibacterial properties and conclusion was made to investigate it as a phytotherapeutic agent to combat infectious agents (Ogbunugafor *et al.,* 2011; Patel, 2011).

Many adult worms of *Prohemistomum vivax* were observed in rats stool from 2nd day of moringa treatment, also, no worms were recovered at the 5th day of treatment with moringa and the duodenal sections in treated rats shows normal histological structure, with only a few hemorrhage, mild villi atrophy with an increased in goblet cell numbers (Figures 31-38). The present results agree with Thilza et al. (2010) who reported that extract from moringa leaf acts as anti-helminthic activity, antimicrobial activity, detoxifier, immune booster and anti – parasitic activity. The study supports and validates the traditional use of moringa leaf extract and further confirms that the oleic acid present in Moringa may also contributes to the traditionally claimed anthelmintic activity .

References

1. Anwer F, Latif S, Ashraf M, Gilani A.H. Moringa oleifera: a food plant with multiple medicinal uses. Phytother. Res. 2007; 21: 17–25.
2. Azim MA. On the intestinal helminthes of dogs in Egypt, J. Egypt. Med. Ass. 1938; 21(3): 1-5.

3. Bancroft JD, Stevens A. Theory and Practice of Histological Technique. 3rd Ed. Churchill Livingstone. Edinburgh, London 1990.
4. Bharali, R., Tabassum, J., Azad, M.R., 2003. Chemomodulatory effect of Moringa oleifera Lam on hepatic carcinogen metabolizing enzymes, antioxidant parameters and skin papillomagenesis in mice. Asian Pac. J. Cancer Prev. 2003; 4: 131–139.
5. Bowman, DD et al (2003) Feline clinical parasitology. Iowa University Press, Iowa. pp:89-91
6. El-Naffar, M.K. and El-Shahawi G.A.Z. (1986): Studies on the metacercariae of the Nile fishes at El-Mini Province, A.R. Egypt. Assiut Vet. Med. J. 15. (30) 46:58.
7. El-Naffar, M.K.; Saoud, M.F. and Hassan, I.M. (1985): Role played by fish in transmitting some trematodes of dogs and cats at Asswan Province, A.E. Egypt. Assiut Vet. Med. J. 14, (27)56:72.
8. Fahmy, M.A.; Mandour, A.M. and El-Naffar, M.K. (1976): Successful infection of dogs and cats by *Prohemistomum vivax* sonsino, 1893 and *Haplorchis yokogawi* Katsuta, 1922. Journal of the Egyptian Society of Parasitology, 6: 77-82.
9. Fahmy, M.A.M. and Selim, M.K. (1959): Studies on some trematode parasites of dogs in Egypt with special reference to the role played by fish in their transmission Z. Parasitenkunde, 19: 3-13.
10. Fayek, S.A.; Abd EL-Wahab, T.M. and Raef, A.M. (1999): Nematode parasites of crayfish (*Procambarus clarkii*) at Kafr EL-Sheikh Province. Alex. J. Vet. Science, 15(1): 79-88.
11. Fayek, S.A.; Raef, A.M. and Abd El-Wahab, T.M. (1997): Some studies on helminth parasites encystation in *Penaeus seliferus* (white shrimp) at Port Said, Egypt. Zag. Vet. J., 25(3): 29-33.
12. Graczyk, T.K. and Fried, B. (1998): Echinostomiasis a common but forgotten food-borne disease. Am. J. Trop. Med. Hyg., 58(4): 501-504.
13. Hassan E, Soliman M, Ghobashy A. Some factors affecting metacercarial infections in *Oreochromis niloticus* collected from brackish water, Ismailia, Egypt. Egypt. Acad. J. Biolog. Sci., 2012; 4(1): 29 -37
14. Hernandez EM, Constantino C, Gracia M, Osorlo SO. Gill lesions in common carp, Cyprinus carpio in Mexico due to the metacercariae of *Centocestus formosonus*. J. Fish Dis., 1998; 21: 229-232.
15. Kakengi AMV, Kaijage JT, Sarwatt SV, Mutayoba SK, Shem MN, Fujihara T. Effect of *Moringa oleifera* leaf meal as a substitute for sunflower seed meal on performance of laying hens in Tanzania. In Livestock Research and Rural Development, 2007; 19(8).
16. Khalifa R, El-Naffar MK, Arafa MS. Studies on heterophid cercariae from Assiut Province. 1- Notes on the life cycle of *Haplorchis pumilio* (Looss, 1896) Trematoda: Heterophyidae) with a discussion on previously described forms. Acta parasitol. Polonica, 1977; 25 (3): 25: 38.
17. Khalil AI. Biocemical and pathological changes in Clarias lazera (cat fish) naturally infected with metacercariae parasites in Gharbiah province. Ph.D. Thesis. Faculty of Science, Tanta Univesity, Tanta, Egypt 1987.
18. Kumar A, Pari L. Antioxidant action of Moringa oleifera Lam (drumstick) against antitubercular drugs induced lipid peroxidation in rats. J. Med. Food, 2003; 6: 255–259.
19. Mahajan SG, Mehta AA. Effect of Moringa oleifera Lam. Seed extract on ovalbumin-induced airway inflammation in guinea pigs. Inhal. Toxicol., 2008; 20: 897–909.
20. MAKKAR HPS, BECKER K. Nutritional value and an nutritional components of whole and extracted *Moringa oleifera* leaves. In *Animal Feed Science and Technology*, 1996; 63: 211-228.
21. Nasr M. The occurrence of *Prohemistomum vivax* infections in man with redescription of the parasite. Lab. and Med. Prog., 1941; 2: 135–149.
22. Noga EN. Fish diseases (diagnosis and treatment) Blackwell Science Ltd., U.S.A. 2000; 378.
23. Ogbunugafor HA, Eneh FU, Ozumba AN, Igwo-Ezikpe MN, Okpuzor J, Igwilo IO, Adenekan SO, Onyekwelu OA. Physico-chemical and anti-oxidant properties of *Moringa oleifera* seed oil. In *Pakistan Journal of Nutrition,* 2011; 10: 409-414.
24. Olugbemi TS, Mutayoba SK, Lekule FP. Effect of Moringa (*Moringa oleifera*) Inclusion in Cassava based diets to broiler chickens. In *International Journal of Poultry Science,* 2010; 9 (4): 363-367.
25. Park CW, Kim JS, Joo HS, Kim J. A human case of Clinostomum complanatum infection in Korea. Korean J Parasitol 2009; 47: 401-404.
26. PATEL JP. Antibacterial activity of methanolic and acetone extract of some medicinal plants used in India folklore. In *International Journal of Phytomedicine,* 2011; 3: 261-269.
27. Raef AM. Role of marine fish in transmission of some parasites to animals and birds. Ph.D.

Thesis, Parasitology, Fac. Vet. Med. Zagazig. University1994.
28. Raef AM, Mohamed AA, Abd El-Maksoud, SA. Role of blue crabs "*Callinectes sabidus*" in transmission of *Paragonimus* (lung fluke) to dog at Sharkia fish markets. Beni-Suef Vet. Med. J., 1999: 9(3-A): 299-309.
29. Raef AM, Mohamed AA, Mohamed MEM, Abd El-Maksoud SA. Further studies on the role of crayfish (*Procambarus clarki*) in transmission of some zoonotic parasites in East Delta. The third international Scientific Conference, Mansoura, Fac. Vet. Med., Mansoura Univ., 2003: 151-158.
30. Rifaat MA, Salem SA, El-Kholy SI, Hegazi MM, Yousef M.El-M. Studies on the incidence of *Heterophyes heterophyes* in Dakahlia Governorate. Journal of the Egyptian society of Parasitology, 1980; 10 (2): 369-373.
31. Saba SER. Some studies on parasites encystations of fresh water fishes. Ph.D. thesis, Parasitology, Fac. Vet. Med. Zagazig. Univ. 2004.
32. Salgado-Maldonado G, Pineda-López RF. The Asian fish tapeworm *Bothriocephalus acheilognathi*: A potential threat to native freshwater fish species in Mexico. Biol. Invasions, 2003; 5: 261-268.
33. Scholz T. Parasites in cultured and feral fish. Vet. Parasitol., 1999; 84(3-4): 317-335.
34. Shalaby SIA. Studies on the role played by some Nile fishes in transmitting trematodes worms to dogs. M.V.Sc. Thesis, Parasitology, Fac. Vet. Med. Cairo Univ. 1982.
35. Shalaby SIA. Further studies on the role played by some cat-fish in transmitting some trematodes to fish eating mammals with special reference to the morphobiology of *Mesostephanus appendiculatus*. Ph.D. Thesis, Parasitology, Fac. Vet. Med. Cairo Univ. 1985.
36. Shalaby SIA. Communicable parasites from Nile fishes. II. Intestinal pathology in final host induced by Haplorchid flukes. Egypt. J. Comp. Pathol. and Clin. Pathol., 1993; 6(1): 189-197.
37. Tadros G, El. and El-Mokadem EEA. Observations on heterophyids affecting man in Egypt. Bull. Zool. Soc. Egypt, 1983; 33: 107-111.
38. Taher GA. Some studies on metacercarial infection in oreochromis niloticus in assiut governorate and their role in transmission of some trematodes to dogs. Ass. Univ. Bull. Environ. Res., 2009; 12: 1.
39. Tantawy EA. Muscular parasites in market fishes. M.V.Sc. thesis, Hygiene and control of meat, fish and their products and animal by products, Fac. Vet. Med. Cairo. Univ. 1993.
40. Thilza I, Sanni S, Zakari A, Muhammed T, Musa B. In vitro antimicrobial of water extract of *Moringa oleifera* leaf stalk on bacterial normally implicated in eye disease. Acad. Aren. 2010; 2:80-83.
41. Thurston G, Murphy TJ, Baluk P, Lindsey JR, McDonald DM. Angiogenesis in mice with chronic airway inflammation: Straindependent differences. Am. J. Pathol., 1998; 153:1099–1112.
42. Williams HH, Jones, A. Marine helminthes and human health. Commonu. Inst. Helminthol. Misc. Publ. 1976; 3: 47.
43. World Health Organization. Control of foodborne trematodes infections. Geneva: WHO; 1995; 1-107.
44. Yooyen T, Wongsawad C, Kumchoo K, Chaiyapo M. A new record of Clinostomum philippinensis (Valasquez, 1959) in Trichogaster microlepis (Gunther, 1861) from Bung Borapet, Nakhon Sawan, Thailand. Southeast Asian J Trop Med Public Health 2006; 37: 99-103.

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