

Infestation of Some Marine Fish Species with Red Worm *Philometra*

إصابة بعض فصائل الأسماك البحرية بالديدان الحمراء "الفيلومترا"

Mohamed A. H.¹, Hassan, M. A.² and Mahmoud M. A.³

عبد المحسن حسن محمد عوض، محمد عبد العزيز حسن، ومحمود علي محمود

¹Department of Fish Diseases, Animal Health Research Institute, Dokki, Giza, Egypt

²Fisheries Research Center, Eastern Province, Kingdom of Saudi Arabia

³Faculty of Veterinary Medicine, Cairo University

Abstract: Over a two-year period, November 2006 to October 2008, the distribution of *philometra* sp. among host tissues was investigated in 10 host species collected from some coastal waters areas of the Arabian Gulf, Kingdom of Saudi Arabia. The examined fish species were: *Epinephelus chlorostigma*, *Epinephelus tauvina*, *Johnius maculates*, *Lethrinus nebulosus*, *Lutjanus ehrenbergi*, *Psettodes erumei*, *Scomberomorus commerson*, *Siganus canaliculatus*, *Trichiurus lepturus* and *Tylosurus crocodiles*. The overall percent of *philometra* sp. infestation among the examined fish species reached 9.5%. However, the infestation percentages in *E. tauvina*, *L. nebulosus* and *E. chlorostigma*, reached 24.13, 21.87 and 20 respectively, whereas in the rest of the examined fish species it was 0%. Generally, *philometra* sp. infestation rates showed seasonal variations, peaked in summer (25.92%), followed by the spring season (24.48%), winter season (21.21%) and autumn season (13.88%). The distribution of *philometra* sp. in tissues of infested fish species revealed its significant existence in the abdominal cavity and gonads. However, infestation rates in muscles and fin tissue were markedly and significantly low. Regarding histo-pathological alterations, infected ovaries appeared black, shrunken and changed to fibrous tissue. Degenerative changes and inflammatory cell proliferation were observed, however, in cases of chronic infestation, the dead parasite in the ovarian tissue initiated strong tissue response and proliferated connective tissue reaction as well as hyaline degeneration. Consequently, reduced in the volume of the affected ovaries and finally damage to the reproductive system were frequently seen.

Keywords: Fish, marine water, Arabian Gulf, *Philometra*, seasonal variations, infestation, pathology.

المستخلص: تم رصد مدى انتشار دودة الفيلومترا في أنسجة لعشرة فصائل من الأسماك البحرية تم تجميعها من بعض مناطق المياه الساحلية للخليج العربي بالملكة العربية السعودية وهي أسماك السممان، الهامور، الشماهي، الشعري، النيسرة، الخوفع، الكنعن، الصايف، السيف راندو والحاقول. وقد بلغت نسبة الإصابة الكلية في الأسماك التي تم فحصها 9,5%؛ حيث بلغت نسبة الإصابة في أسماك الهامور والشعري والسممان 24,13%، و 21,87% و 20% على التوالي، بينما لم يتم تسجيل أي إصابات في بقية الأنواع من الأسماك التي تم فحصها. سجلت أعلى إصابة بدودة الفيلومترا في فصل الصيف (25,92%) يليه فصل الربيع (44,48%) ثم فصل الشتاء (21,21%) ثم فصل الخريف (13,88%). تركزت الإصابة بدودة الفيلومترا في التجويف البطني والمبايض للأسماك المصابة، بينما تواجدت بصورة أقل في الزعانف والعضلات. أما فيما يتعلق بالتغيرات الهستوباثولوجية؛ فقد ظهرت المبايض سوداء اللون،

منكمشة وأصبحت ذات نسيج ليفي، مع وجود انتشار للخلايا الإلتهابية وتغيرات أخرى انجلائية. أما في حالة الإصابة المزمنة؛ فإن تواجد الطفيل الميت في أنسجة المبيض قد يؤدي إلى حدوث رد فعلي نسيجي قوي يؤثر سلباً على حجم وكفاءة المبيض، والذي يؤدي بدوره إلى فقد لوظائف الجهاز التناسلي للأسماك المصابة.

كلمات مدخلية: أسماك، المياه البحرية، الخليج العربي، دودة الفيلومترا، التغيرات الموسمية، نسبة الإصابة، التغيرات النسيجية.

INTRODUCTION

Philometrids are large filamentous, viviparous nematodes living in the fish body cavity, between fin rays, in the subcutaneous tissue and in gonads (Grbada, 1991; Hesp, *et al.* 2002 ; Moravec, *et al.* 2003 ; Clarke, *et al.* 2006 and Burak, 2007). The living parasite survives by sucking the blood of affected fish, so it is commonly called the red worm (Oliva, *et al.* 1992 and Burak, 2007). Females produce juveniles which exit the host and must be eaten by a cyclopoid copepod. Fish become infested by feeding on the copepods which harbor third stage larvae (L_3) (Molnar, 1966 b and Moravec, 1980 and Hesp, *et al.* 2002).

Philometra sp. has been reported to infest the body cavity, swim bladder, gonads, subcutaneous tissues, fillets, ocular cavity and fins of both freshwater and marine fish in different parts of the world (Obiekezie, *et al.* 1992; Vidal-Martinez, *et al.* 1995 ; Gelnar, *et al.* 1997; Moravec, *et al.* 1997; Pazooki and Molnar , 1998 ; Moravec, *et al.* 2003 and Quiazon, *et al.* 2008).

The adult female worms are large and red, so their occurrence in fish flesh or body cavities of their hosts often cause rejection of such parasitized fish. The gonad-dwelling species are of considerable economic importance as they may cause partial or total sterility of the host in heavy infestation, thus reducing the affected fish population (Grbada, 1991; Moravec, *et al.* 1997 and Burak., 2007). Some species of *philometra* sp. may disrupt the normal functioning of the swim-bladder, causing loss of equilibrium. A case of an adult female dracunculoid *philometra* sp. which invaded a puncture wound in a fisherman's hand while he was filleting an infected carangidae fish, *Caranx melampygus*, was reported in Hawaii (Deardorff, *et al.* 1986).

No former records of *philometra* sp. in the Arabian Gulf fishes are available; therefore, the present study was designed to examine on *philometra* sp. which may infest the fish of the Arabian Gulf on the Saudi coast: to detect the percentage and the intensity of infestation, record seasonal variation of infestation and lastly investigate the histo-pathological alterations which may be produced by this parasite.

MATERIALS AND METHODS

Over a two-year period, November 2006 to October 2008, a total of 400 fish specimens, representing 10 species: *E. chlorostigma*, *E. tauvina*, *J. maculates*, *L. nebulosus*, *L. ehrenbergi*, *P. erumei*, *S. commerson*, *S. canaliculatus*, *T. lepturus* and *T. crocodiles* (Table 1) were randomly collected from various locals of the Arabian Gulf at Qatif in the Eastern Province of Saudi Arabia (Figure 1), in different seasons. Fish specimens were measured to the nearest cm of total length and weighed to the nearest gm. Seasonal infestation and correlation between fish size and infestation were studied.

Collected fish were dissected and their different tissues: fins, subcutaneous tissues, musculature, body cavity, gonads, swim bladder and ocular cavity were examined for *philometra* sp. infestation (Roberts, 2001). The search was done macroscopically so *philometra* sp. (red worm) were easily discerned in affected tissue.

Recovered parasites were removed, counted and fixed in 10% formaldehyde solution ora mixture of 70% ethanol and 5% glycerol. Specimens were stained using Acetic-Carmine, dehydrated and mounted in canadabalsam. Sections of affected tissues were appropriately fixed, embedded in paraffin wax, cut to a thickness of 5-7 μ m and stained with haematoxylin and eosin for histo-pathological studies (Roberts, 2001).

Table 1. List of fish species studied.

Scientific name	English name	Family
		Scientific name
<i>Epinephelus tauvina</i>	Greasy grouper	Serranidae
<i>Lethrinus nebulosus</i>	Spangled emperor	Lethrinidae
<i>Epinephelus chlorostigma</i>	Brown-spotted grouper	Serranidae
<i>Johnius maculatus</i>	Blotched croaker	Sciaenidae
<i>Lutjanus ehrenbergi</i>	Ehrenberg's snapper	Lutjanidae
<i>Psettodes erumei</i>	Indian Spiny turbot	Psettodidae
<i>Scomberomorus commerson</i>	Narrowbarred Spanish mackerel	Scombtidae
<i>Siganus canaliculatus</i>	Whitespotted spinfoot	Siganidae
<i>Trichiurus lepturus</i>	Cutlassfishes and hairtailfishes	Trichiuridae
<i>Tylosurus crocodilus</i>	Gar fish	Belonidae

**Fig. 1.** Map showing the study area (Qatif area).

RESULTS

In this study, philometra sp. was detected in infested fish species as a bloody-red to dark red colored filamentous thread (Figure 2-A), cylindrical, of an average 30 cm in length and 1 mm in width. Males were not recovered in this study. The parasite is characterized by the

presence of conspicuously large, crescent-shaped, fleshy cephalic papillae of the external circle and small, sub-terminal papilla-like projections on the caudal end; however, the mouth opening is small and surrounded with papillae, the oesophagus has an anterior muscular part and posterior glandular part (Figure 2-B). The intestine is a straight, dark-brown tube with the absence of an anus, the vulva

cannot be detected and the larvae were observed in the uterus or abdominal cavity of the worm (Figure 2-C). Blood elements were seen in the cross-section of the intestine of philometra worms.

Table (2) demonstrates the overall percentage of philometra sp. infestation among examined fish species reached 9.5%. However, the infestation percentages in *E. tauvina*, *L. nebulosus* and *E. chlorostigma*, reached 24.13, 21.87 and 20 respectively. In the remainder of the examined fish species: *J. maculates*, *L. ehrenbergi*, *P. erumei*, *S. commerson*, *S. analiculatus*, *T. lepturus* and *T. crocodiles* were 0 %.

A relationship was found between the size of the fish and its infestation with philometra sp. Table (2) records the infestation detected in *E. chlorostigma*, *E. tauvina* and *L. nebulosus* at more than 68 cm, 63 cm and 45 cm respectively.

Table (3) clarifies the seasonal variations of philometra sp. infestation rates. Generally, infestation peaked during the Summer season (25.92%), followed by the Spring season (24.48%), Winter (21.21%) and Autumn (13.88%) with some variations among the three infested fish species: *E. tauvina*, *L. nebulosus* and *E. chlorostigma*.

Clinically, philometra sp. infestation in the fins of *E. tauvina* appeared as a dark red filamentous thread between the fin rays of the pectoral fins (Figure 3). However, fish infested with philometra sp. in their abdominal cavity, gonads and musculature showed no gross abnormalities. Data recorded in Table (4) reveals philometra sp.

occurred mostly in the abdominal cavity (Figure 4) and gonads (Figures 5,6,7) of *E. chlorostigma*, *E. tauvina*, and *L. nebulosus*, while light fin and musculature infestations were recorded in *E. tauvina* (Figure 3) and *L. nebulosus* (Figure 8) respectively. The coiled form of philometra sp. were frequently observed within the abdominal cavity and ovarian tissues with a tendency to form several coils.

Histologically, philometra sp. was detected macroscopically in the ovarian tissue of infested fish as dark red filamentous threads; however, microscopically, the infected ovaries showed an embedded parasite with variable degrees of tissue reaction depending upon the number and condition of the nematodes as well as the duration of infestation. Regarding histopathological alterations, degenerative changes and inflammatory cell proliferation were generally seen. However, masses of developmental stages of philometra sp. surrounding the ovarian oocytes (Figure 9) with some of the secondary oocytes free from the ova (Figure 10) were commonly seen in most infested fish species. In more advanced cases of chronic ovarian infestation, the ovaries changed grossly to dark shrunken fibrous tissue. Microscopically, the dead parasite appeared as a black remnant of an irregular area surrounded by massive proliferation of fibrous connective tissue and inflammatory cells accompanied by degeneration, diffuse necrosis of the follicles as well as the centers of the oocysts (Figures 11 & 12). The affected muscular tissue, in general, showed an inflammatory reaction surrounding the adult philometra (Figure 13).

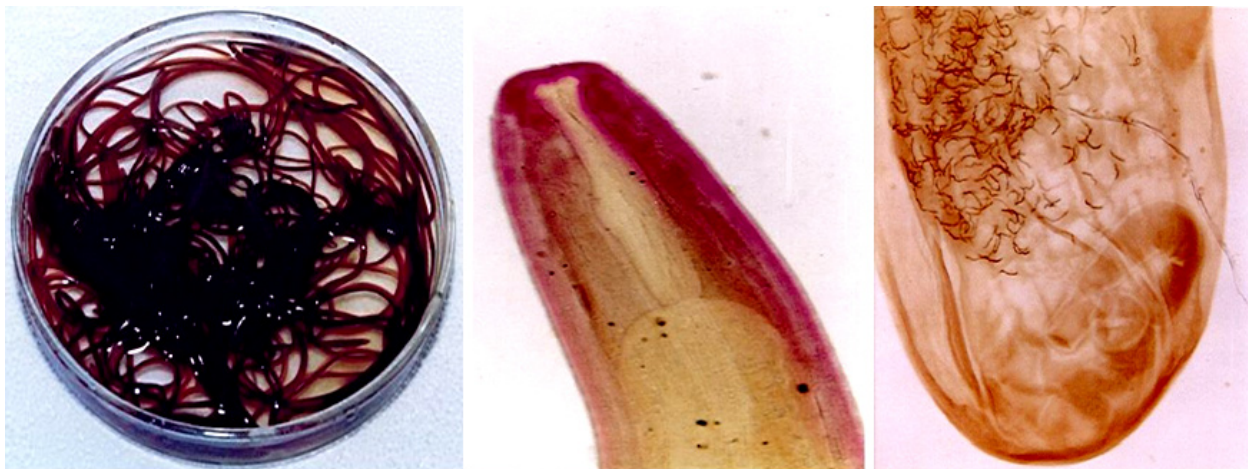


Fig. 2. (A-C)

A. Mass of mature philometra, **B.** Anterior of mature worm, **C.** Posterior of mature worm

Table 2. Occurrence of *Philometra* sp. in local fish species from the Arabian Gulf.

Species	Length (cm)	No. Exam.	No. Infect.	%
<i>Epinephelus tauvina</i>	26 - 92	58	14*	24.13
<i>Lethrinus nebulosus</i>	25.5 - 69	64	14**	21.87
<i>Epinephelus chlorostigma</i>	30 - 95	50	10***	20
<i>Johnius maculatus</i>	24 - 58	34	0	0
<i>Lutjanus ehrenbergi</i>	16 - 36	32	0	0
<i>Psettodes erumei</i>	32 - 63	36	0	0
<i>Scomberomorus commerson</i>	52 - 73	30	0	0
<i>Siganus canaliculatus</i>	18 - 35	32	0	0
<i>Trichiurus lepturus</i>	59 - 79	30	0	0
<i>Tylosurus crocodilus</i>	67 - 99	34	0	0
Total		400	38	9.5

* *Epinephelus tauvina* 63 > cm in length

** *Lethrinus nebulosus* 45 > cm in length

*** *Epinephelus chlorostigma* 68 > cm in length

Table 3. Seasonal incidence of *Philometra* sp. infestation.

	Winter			Spring			Summer			Autumn		
	No. Exam.	No. Infect.	%	No. Exam.	No. Infect.	%	No. Exam.	No. Infect.	%	No. Exam.	No. Infect.	%
<i>Epinephelus chlorostigma</i>	10	2	20	14	4	28.57	14	2	14.28	12	2	16.66
<i>Epinephelus tauvina</i>	11	2	18.18	17	4	23.52	18	6	33.33	12	2	16.66
<i>Lethrinus nebulosus</i>	12	3	25	18	4	22.22	22	6	27.27	12	1	8.33
Total	33	7	21.21	49	12	24.48	54	14	25.92	36	5	13.88

Table 4. Infestation sites of *Philometra* sp.

Fish species	Muscles	Abdominal cavity	Gonads	Fins
<i>Epinephelus chlorostigma</i>	-	+++	+++	-
<i>Epinephelus tauvina</i>	-	+++	+++	+
<i>Lethrinus nebulosus</i>	+	+++	+++	-

**Fig. 3.** *Philometra* sp. in the pectoral fin of *Epinephelus tauvina*

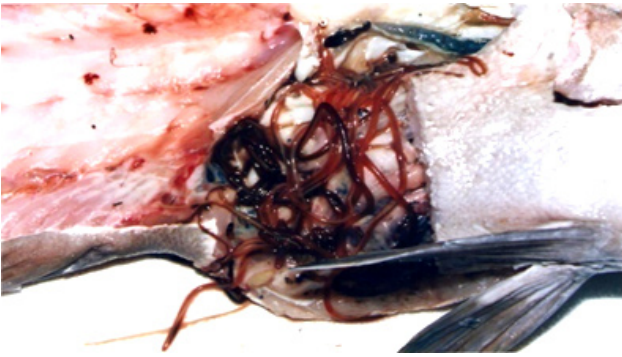


Fig. 4. Masses of mature philometra in the abdominal cavity of *Epinephelus chloristigma*.

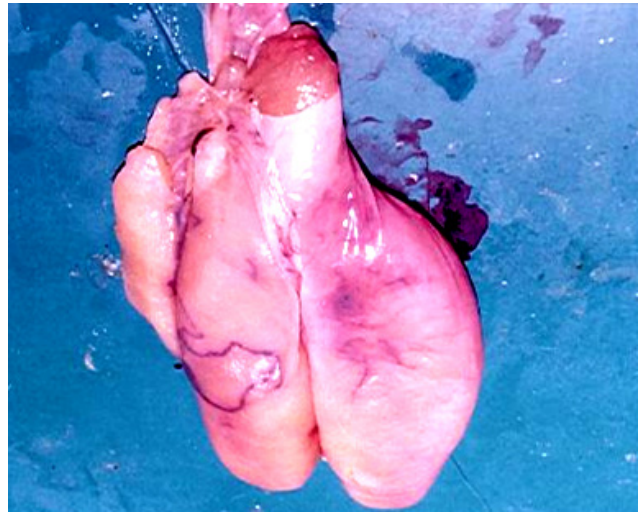


Fig. 7. Gonads of *Epinephelus chloristigma* infected with philometra.

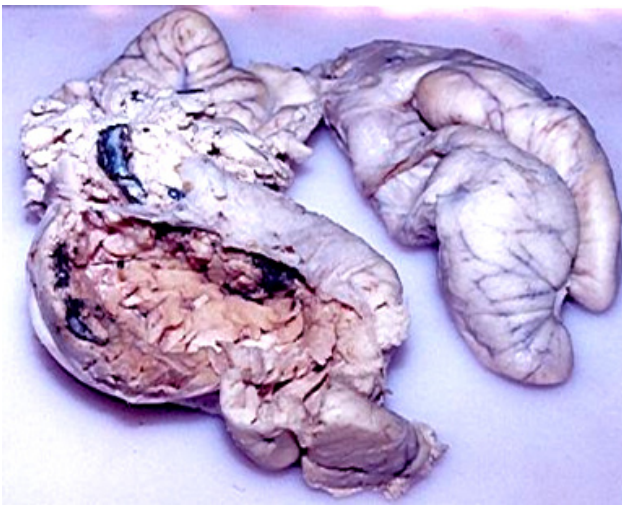


Fig. 5. Gonads of *Lethrinus nebulosus* infected with philometra.

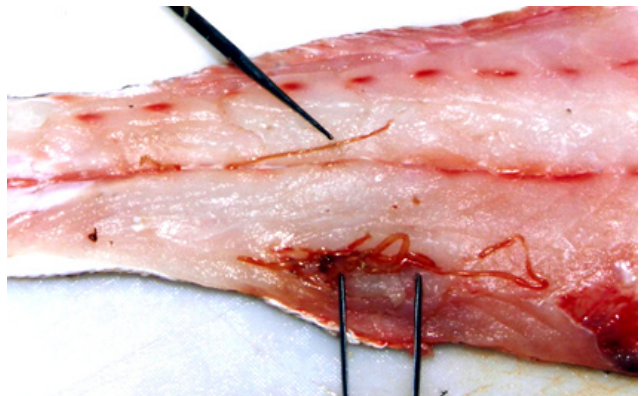


Fig. 8. *Philometra* sp. Embedded in the muscle of *Lethrinus nebulosus*.



Fig. 6. Gonads of *Epinephelus tauvina* infected with philometra.

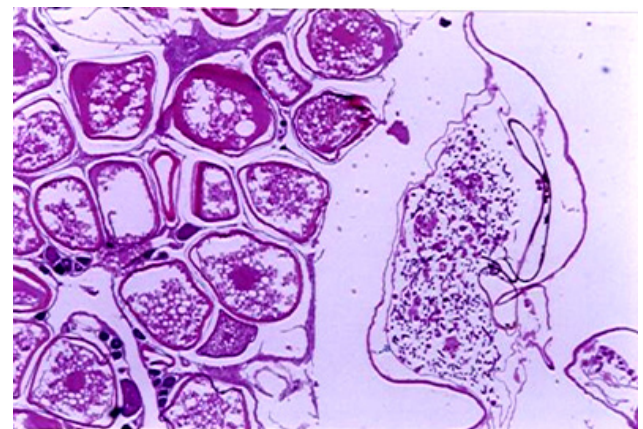


Fig. 9. Ovary of *Epinephelus tauvina* showing masses of nematodes surrounding the oocysts with developmental stages of the parasites (H & E X 40).

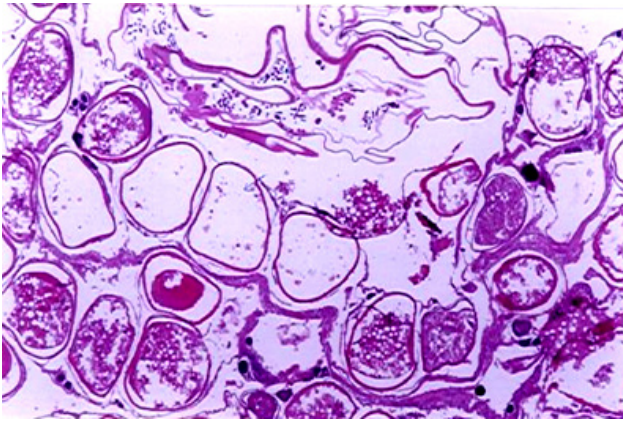


Fig. 10. Ovary of *Epinephelus tauvina* showing clusters of nematodes and some of The secondary oocysts were free from ova (H & E X 40).

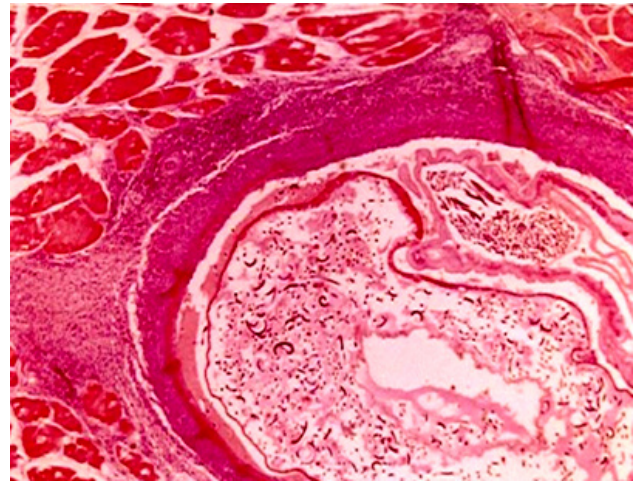


Fig. 13. Muscle of *Lethrinus nebulosus* showing inflammatory reaction surrounding an adult philometra.

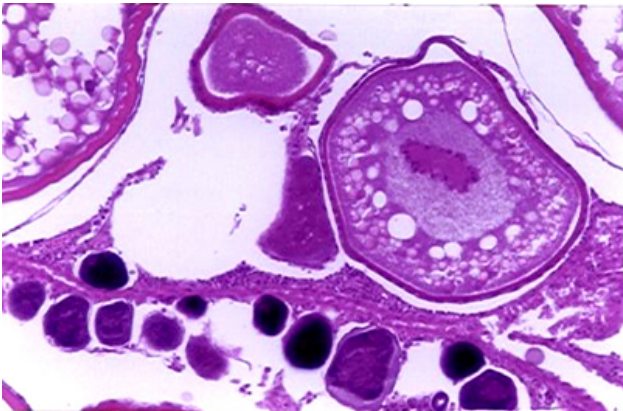


Fig. 11. Ovary of *Epinephelus chloris chloristigma* infected with philometra showing degeneration and necrosis in the center of oocysts (H & E X 400).

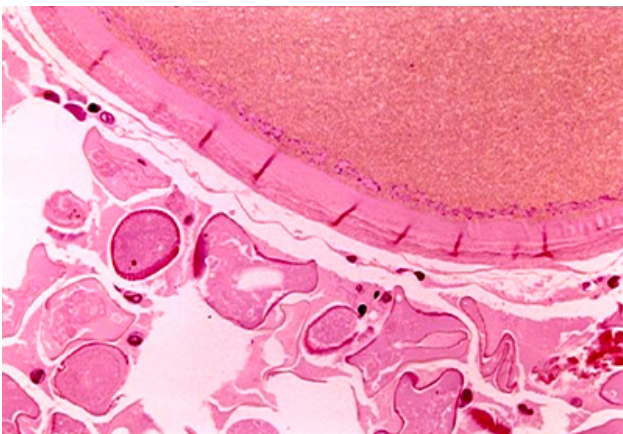


Fig. 12. Ovary of *Lethrinus nebulosus* infected with adult philometra showing degeneration of oocysts.

DISCUSSION

Philometrids are filamentous, large viviparous nematodes living in the fish body cavity, between fin rays, in the subcutaneous tissue and in gonads (Grbada, 1991; Hesp, *et al.* 2002; Moravec, *et al.* 2003; Clarke, *et al.* 2006 and Burak, 2007). *Philometra* sp. was detected in this study as a bloody-red to dark red colored filamentous thread, (Figure 2-A) cylindrical, of an average 30 cm in length and 1 mm in width. Males could not be recovered in this study, supporting the same findings of (Grbada, 1991) and (Hesp, *et al.* 2002). The parasite is characterized by the presence of conspicuously large, crescent-shaped, fleshy cephalic papillae of the external circle and small sub-terminal papilla-like projections on the caudal end; however, the mouth opening is small and surrounded with papillae while the oesophagus has an anterior muscular part and a posterior glandular part (Figure 2-B). The intestine is a straight dark-brown tube with the absence of anus and the vulva cannot be detected, larvae were observed in the uterus or abdominal cavity of the worm (Figure 2-C). Our specimens resemble those described by Yamaguti (1935), however, the average specimen length recorded by Yamaguti was 10 cm. In this regard, Knight (1984) reported that the measurements of philometrids could be different in various infested fish species. Blood elements were observed in the

cross-section of the intestine of the philometra worms, confirming data that the living parasite survives on suckling the blood affected fish (Oliva, *et al.* 1992, Oliva, *et al.* 1996, Hesp, *et al.* 2002 and Clarke, *et al.* 2006), This diversion of nutrients to the parasite may exacerbate the impact of the worms on ovarian tissue (Clarke, *et al.* 2006) .

Philometra sp. has been reported to infest the body cavity, swim bladder, gonads, subcutaneous tissues, fillets, ocular cavity and fins of both freshwater and marine fish in different parts of the world (Obiekezie, *et al.* 1992; Vidal-Martinez, *et al.* 1995 ; Gelnar, *et al.* 1997; Moravec, *et al.* 1997; Pazooki and Molnar, 1998; Moravec, *et al.* 2003 and Quiazon, *et al.* 2008). However, Moravec, *et al.* (1997) reported that *Philometra margolisi*, a nematode parasitizing the gonads, is undoubtedly the most significant parasite affecting the reproduction of the host, endangering *Epinephelus morio* (Red grouper) in aquacultural operations. In addition, Grbada (1991) reported the recovery of *Philometra lateolabrocis* from various marine fish species such as *Lateolabrax Japonies* (Japanese seabass), *Scomberomorus Chinesis* (Chinese Seerfish) and others in Japan, Pakistan and India. However, the gonad-dwelling species are of considerable economic importance as the parasite may cause partial or total sterility of the host in heavy infestations, thus reducing the affected fish population.

The overall percentage of philometra sp. infestation among examined fish species reached 9.5%. However, the infestation percentages in *E. tauvina*, *L. nebulosus* and *E. chlorostigma*, reached 24.13, 21.87 and 20 respectively, whereas the rest of the examined fish species: *J. maculates*, *L. ehrenbergi* , *P. erumei*, *S. commerson*, *S. canaliculatus*, *T. lepturus* and *T. crocodiles* were at 0 % (Table 2). These results proved the presence of significant variations among fish species regarding clinical infestation of philometra sp.

A relationship was found between the size of the fish and its infestation with philometra sp. which was recorded in (Table 2), where infestation was detected in *E. chlorostigma*, *E. tauvina* and *L. nebulosus* more than 68 cm , 63 cm and 45 cm respectively. Our results also support those of

Oliva, *et al.* 1996., Hesp, *et al.* 2002 and Clarke, *et al.* 2006.

Generally, philometra sp. infestation rates showed seasonal variations, peaking in the summer season (25.92%), followed by the Spring season (24.48%), Winter season (21.21%) and Autumn season (13.88%) with some variations among the three infested fish species: *E. tauvina*, *L. nebulosus* and *E. chlorostigma* (Table 3). In this regard, Hesp, *et al.* 2002 and Clarke, *et al.* 2006 and Burak., 2007 reported that the prevalence and intensity of philometra sp. infestation were highest during the Summer and Spring spawning seasons .

Clinically, philometra sp. infestation in the fins of *E. tauvina* appeared as a dark red filamentous thread between the fin rays of the pectoral fins (Figure 3); however, fish infested with philometra sp. in their abdominal cavity, gonads and musculature showed no gross abnormalities. Data recorded in Table (3) revealed that philometra sp. occurred mostly in the abdominal cavity (Figure 4) and gonads (Figures 5,6,7) of *E. chlorostigma*, *E. tauvina*, and *L. nebulosus*, while light fin and musculature infestations were recorded in *E. tauvina* (Figure 3) and *L. nebulosus* (Figure 8), respectively . A coiled form of philometra sp. was frequently seen within the abdominal cavity and ovarian tissues with a tendency to form several coils, confirming the results recorded by (Hesp, *et al.* 2002). Since only *L. nebulosus* showed light flesh infestations, it appears that philometra sp. do not pose a serious danger to the marketability of most commercially important fish species in the area. In this regard, it is interesting to mention visceral infestation of the *E. chlorostigma*, *E. tauvina*, and *L. nebulosus* with philometra sp. may remain undetected in many cases if the fish is prepared whole, as traditionally happens in many Arabic countries. Filleting, however, practiced in western countries, will make the parasites visible, thus reducing the value of affected fish, particularly for export. In this study, the majority of the philometra sp. were isolated from the abdominal cavity, gonads, musculature and pectoral fins, suggesting that these are the preferred sites of infestation (Hesp, *et al.* 2002 ; Moravec, *et al.* 2003 ; Clarke, *et al.* 2006 and Burak, 2007). In this regard, Roberts

(2001) reported that if the adults and larvae of *philometra* sp. are found in the gonads, particularly the ovaries, they can cause severe damage to infested salmonids. However, fish infected by feeding on the copepod intermediate host and nematodes eventually mature within the visceral cavity of the salmonids, where they may cause peritoneal adhesions. Clarke, *et al.* 2006 and Burak., 2007 reported high prevalence, intensity and pathological damage could be important factors in fecundity variation in bluefish.

Histologically, examination of the infested ovarian tissue revealed an embedded parasite with variable degrees of tissue reaction, depending upon the number and condition of existing nematodes as well as duration of infestation. Regarding histopathological alterations, degenerative changes and inflammatory cells proliferation were generally seen as a result of host response to the infestation (Figures 9 & 10). However, in more advanced cases of chronic ovarian infestation, the ovaries changed grossly to dark shrunken fibrous tissue. Microscopically, the dead parasite appeared as a black remnant of irregular area surrounded by massive proliferation of fibrous connective tissue and inflammatory cells, accompanied by degeneration, diffuse necrosis of the follicles as well as the centers of the oocysts (Figures 11 and 12). The affected muscular tissues, in general, showed an inflammatory reaction surrounding the adult *philometra* (Figure 13). Generally, our histopathological findings match those recorded by Ferguson (1989); however, the reduction in gonad size and / or sterility have also been recorded (Williams, 1967; Margolis, 1970, Sindermann, 1990, Hesp, *et al.* 2002 ; Clarke, *et al.* 2006 and Burak., 2007).

CONCLUSION

Philometra species are filamentous, large viviparous nematodes infesting the body cavity, swim bladder, gonads, subcutaneous tissues, fillets, ocular cavity and fins of both freshwater and marine fish in various locations throughout the world. The living parasite survives by sucking blood of affected fish, so it is commonly called the red worm.

The recorded prevalence of *philometra*

species among examined fish species, seasonal distribution, relationship to fish size and pathological alterations to fish body organs, especially ovaries and muscles, proved the clinical importance of *philometra* species and draws attention to the possible reflection of infestation on fecundity and marketability of infested fish, a matter that requires periodical screening of Arabian Gulf fishes to this parasite.

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- Ref. No. (2540)
Rec. 23/11/2009
In-revised form: 24/10/2010