

## **Parasitic Cymothoid Isopods and their Impacts in Commercially Important Fishes From Lake Qarun, Egypt**

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**Abstract :** Cymothoid Isopods are parasitic crustaceans cause serious impacts on the fish population and might lead to fish mortality and consequently economic losses. The present study aims to investigate the prevalence of isopod infestation and their histopathological alterations among three commercially important fish species in lake Qarun, Fayoum province, Egypt. A total of 150 fish samples; *Dicentrarchus labrax* (50) *Solea vulgaris* (50), and *Tilapia zilli* (50) were collected from the lake during the period from March to August 2015. Results revealed the total prevalence of 32.66% with two isopod species identified as *Nerocila orbigny* and *Renocila thresherorum*. Gills affected with *R. thresherorum* showed complete sloughing of the epithelium lining accompanied with severe congestion and edema in the secondary gill lamellae in addition to inflammatory exudates found between the gill filaments. Further studies are needed to estimate the source of isopod infestation problem and to develop a strategy for controlling such problem among Lake Qarun fishes.

**Keywords :** Cymothoidae, Isopods, parasites, Pathology, Lake Qarun.

### **1- Introduction:**

Parasitic diseases especially that caused by external parasites play an important role in the restriction of fisheries and aquaculture development and lead to great problems and economic losses<sup>1</sup>. Many species of commercially important fishes are parasitized by cymothoids (Crustacea: Isopoda; Cymothoidae). They are hermaphrodites and feeding on blood and different tissues of their suitable host<sup>2</sup>. Several isopod species inhabit the buccal cavity, others found in the branchial cavity or mostly attached to the body surface. The isopod life cycle is direct with one host fish (Holoxenic cycle)<sup>3</sup>. From a pathological point of view, parasitic isopods penetrate its mouth part and anchor causing great damage at sites of attachment so can kill the affected fish especially juveniles<sup>4,5,6</sup>.

Little is known about marine isopods in Egypt, particularly those recorded by<sup>7</sup> from Suez canal water, <sup>8,9,10</sup> from Red Sea as well as<sup>11,12</sup> from Mediterranean Sea.

The location that prospected in the present study was lake Qarun, where problem of isopod infestation among fishes was noticed causing fish loss and marketing problems. Consequently, this preliminary study aims to investigate the prevalence of parasitic isopod species among three commercially important fish species in lake Qarun and evaluating their histopathological impacts on the tissues of the affected hosts.

## 2-Material and Methods

### 2.1. Area of investigation and fish sampling (photo.1)

Lake Qarun is a natural saline lakes located in the lowest part of Fayoum Depression in the Western Desert of middle Egypt between the longitudes of 30° 24' & 30° 49' E and latitudes of 29° 24' & 29° 33' N<sup>13</sup>. A total of 150 fish samples belonging to; *Dicentrarchus labrax* (50), *Solea vulgaris* (50) and *Tilapia zilli* (50) were randomly captured with the help of fishermen from different sites along lake Qarun during the period from March to August, 2015. Samples were brought alive in an ice box with their original water to the laboratory of parasitology, Faculty of Veterinary Medicine, Cairo University. Fish samples ranged from 16-28 (22), 10-20 (15) cm and 8-24 (16) cm in total body length for *D. labrax*, *S. vulgaris* and *T. zilli* respectively.

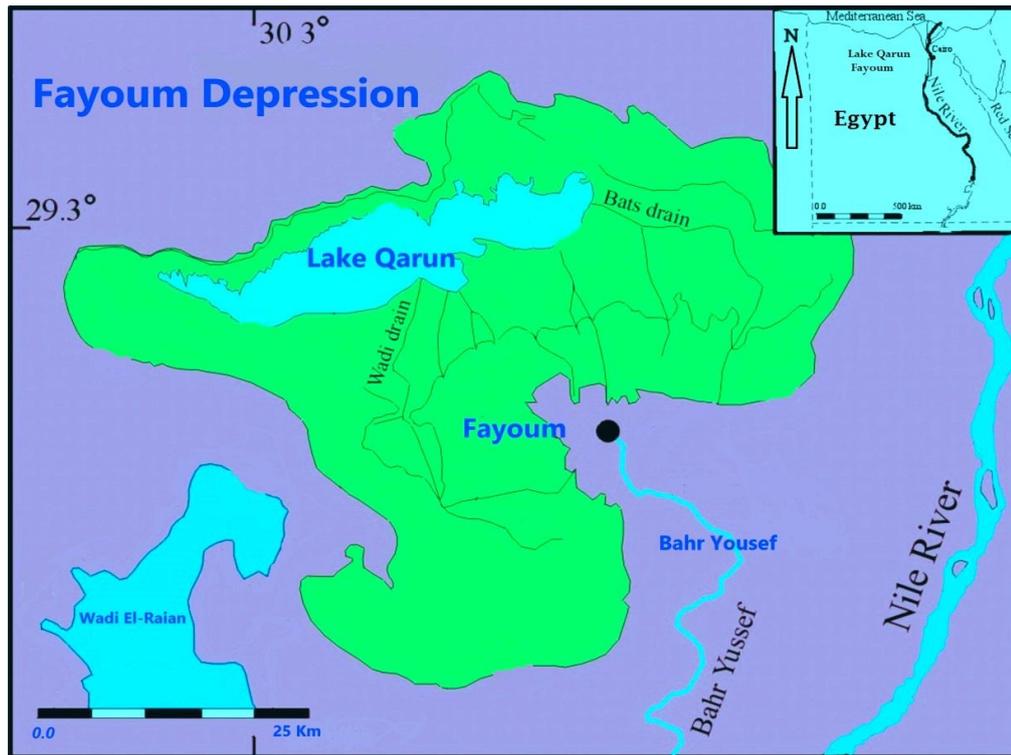


Photo (1): Map showing the investigated area (Lake Qarun)

### 2.2. Clinical examination

Fish under investigation were clinically observed for any abnormal signs and behavior according to<sup>14</sup>.

### 2.3. Parasitological examination

Fish samples were macroscopically examined for any gross lesions and for isopod infestation in body surface including fins, in the branchial cavity and buccal cavity. The detected isopods were removed from the tissues of the infected fish and preserved in 70% ethyl alcohol according to<sup>15</sup>. The isolated isopods were measured in millimeters (mm) with the mean in between brackets, photographed using digital camera (12 megapixels) and morphologically identified according to<sup>16,17</sup>.

### 2.4. Histopathological examination

Gills infested with isopod parasites were fixed in 10% neutral buffered formalin, dehydrated in ascending grades of ethyl alcohol, cleared in xylol then samples were processed by paraffin embedded technique, sectioned at 5 $\mu$  thickness using microtome (**Leica 2135**) and stained with Haematoxylin and Eosin (H&E),<sup>18</sup>. The tissue sections were examined by light microscopy and photographed using camera Olympus XC30 (Tokyo, Japan).

### 3-Results

#### 3.1. Clinical examination

Fish affected with branchial isopod infestation showed signs of respiratory distress manifested by surface swimming with opened mouth and gill cover. Unilateral bulging of the operculum due to the presence of the large sized isopod. In the laboratory fish infested with isopods on their body surface moved in a sluggish manner rubbing their body against the wall of the glass container.

#### 3.2. Parasitological examination

##### 3.2.1. Prevalence:

Out of 150 fish specimens of *Dicentrarchus labrax*(50), *Solea vulgaris* (50) and *Tilapia zilli* ( 50), 49 were found infested with cymothoid isopods (Crustacea: Isopoda; Cymothoidae) with an infestation rate of 32.66%. Two species of cymothoids were detected: *Renocila thresherorum* (females and juveniles) and *Nerocila orbigny* (females) and (table 1).

**Table (1): Prevalence and infestation rate of isopod spp. in the examined fishes.**

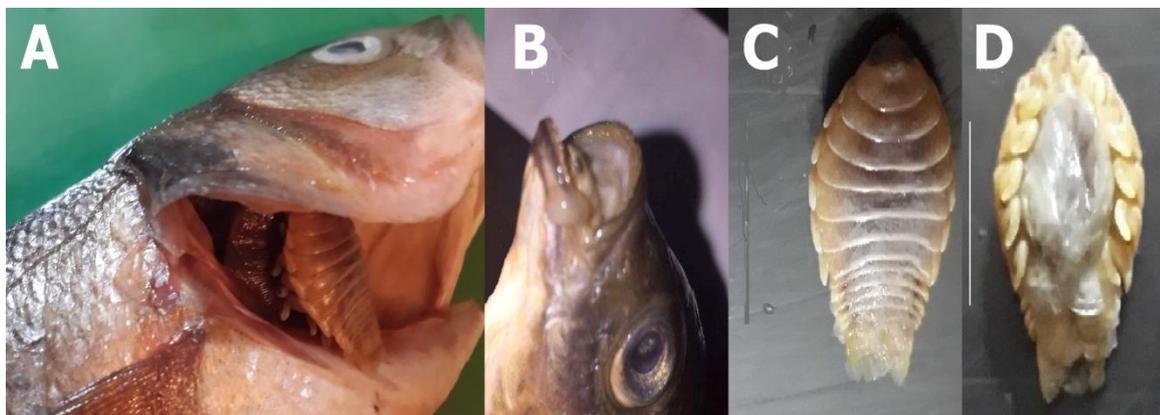
| Fish species           | <i>D. labrax</i><br>No.inf. (%) | <i>S. vulgaris</i><br>No. inf. (%) | <i>T. zilli</i><br>No.inf (%) | Total<br>No.inf. (%) |
|------------------------|---------------------------------|------------------------------------|-------------------------------|----------------------|
| <i>R. thresherorum</i> | 21 (42.00)                      | 0 (0.00)                           | 6 (12.00)                     | 27(18.00)            |
| <i>N. orbigny</i>      | 0 (0.00)                        | 13 (26.00)                         | 9(18.00)                      | 22(14.66)            |
| Total No.inf.(%)       | 21 (24.00)                      | 13(26.00)                          | 15(30.00)                     | 49 (32.66)           |

Total Number of examined fishes =150 (50 from each species).

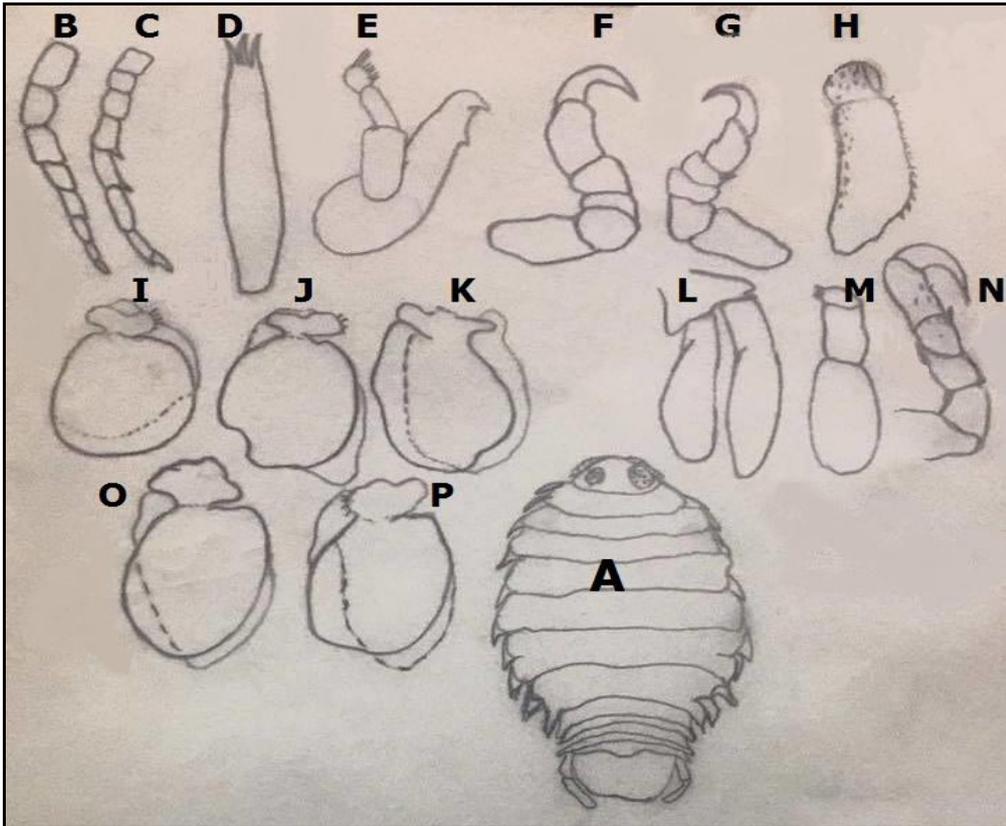
No. inf.: Number infected.

##### 3.2.2. Morphological description of the isolated isopods

##### 2.1- *Renocila thresherorum* ( Williams & Williams, 1980 ) Plate 1 and Fig. 1.



**Plate (1): A: Showing female *R. thresherorum* in gill cavity of *D. labrax*, B: juvenile in the buccal cavity of *T. zilli*, *R. thresherorum*: (C: Dorsal view), (D: ventral view). Scale bar =10mm**



**Figure 1:** A, *R. thresherorum* female dorsal view; B, 1<sup>st</sup> antenna; C, 2<sup>nd</sup> antenna; D, 1<sup>st</sup> maxilla; H, 2<sup>nd</sup> maxilla; E, mandible; F, pereopod1; G, pereopod IV; N, pereopod VII; I, pleopod1; J, pleopod2; K, pleopod3; O, pleopod 4; P, pleopod 5; L, uropod.

Adult females were isolated from the gills of *D. labrax* and *T. zilli*, while juveniles were detected in the buccal cavity of only three *T. zilli* fish.

**Description:** (based on 10 female specimens)

**Body:** Is dark in color with marked concentrated scattered chromatophores on the posterior aspect of segments. It is dorsoventrally compressed measuring 16 -22 (19) mm in total length and 12-16(9) mm in Width.

**Cephalon:** its posterior border is weakly enclosed in pereonite I. Eyes are well developed. The First and second antennae are of 8 articles. The 1<sup>st</sup> antenna is barely reaching the anterior border of pereonite I while the 2<sup>nd</sup> one is barely also and reaching the anterior quarter of pereonite.

**Pereon:** Pereonites I and V are the longest while II and III are the shortest. Pereonites IV-VI are nearly subequal in length. Pereonites V and VI are the widest. Pereonite VII often covers the lateral margins of pleonite I. In dorsal aspect, coxae II and III are about reaching the posterior margins of their pereonites. Coxae of IV-VI I Posterior angles and that of IV are rounded while of V-VII are subacute. Pereopods, with posteriorly are increasing gradually in length. Pereopod with one or two spines on outside of merus Pereopods V-VII are with small spines on inside border of propus and carpus, absent on V-VI in larger specimens. The Posterior pereopods are more spinose than the anterior pereopods.

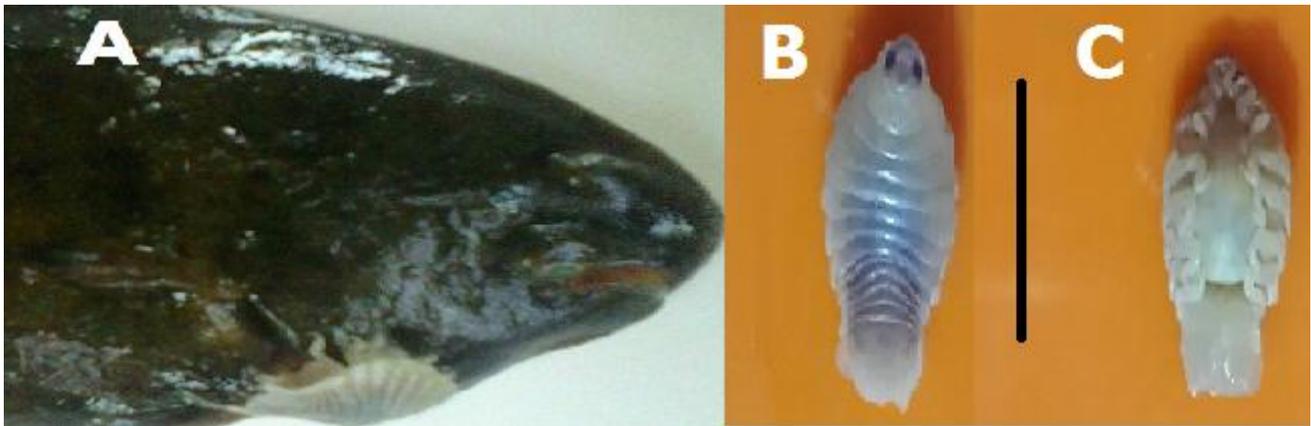
**Pleon:** Pleonites are subequal in length and width. Uropods extended beyond the posterior margin of pleotelson. Pleotelson posterior margin is rounded with its length subequal to width. Uropodal endopod is ovate while exopod is elongated and longer than endopod.

**2.2-Nerocila orbigny (Guérin-Meneville, 1829-1832) Plate 2 and Fig2.**

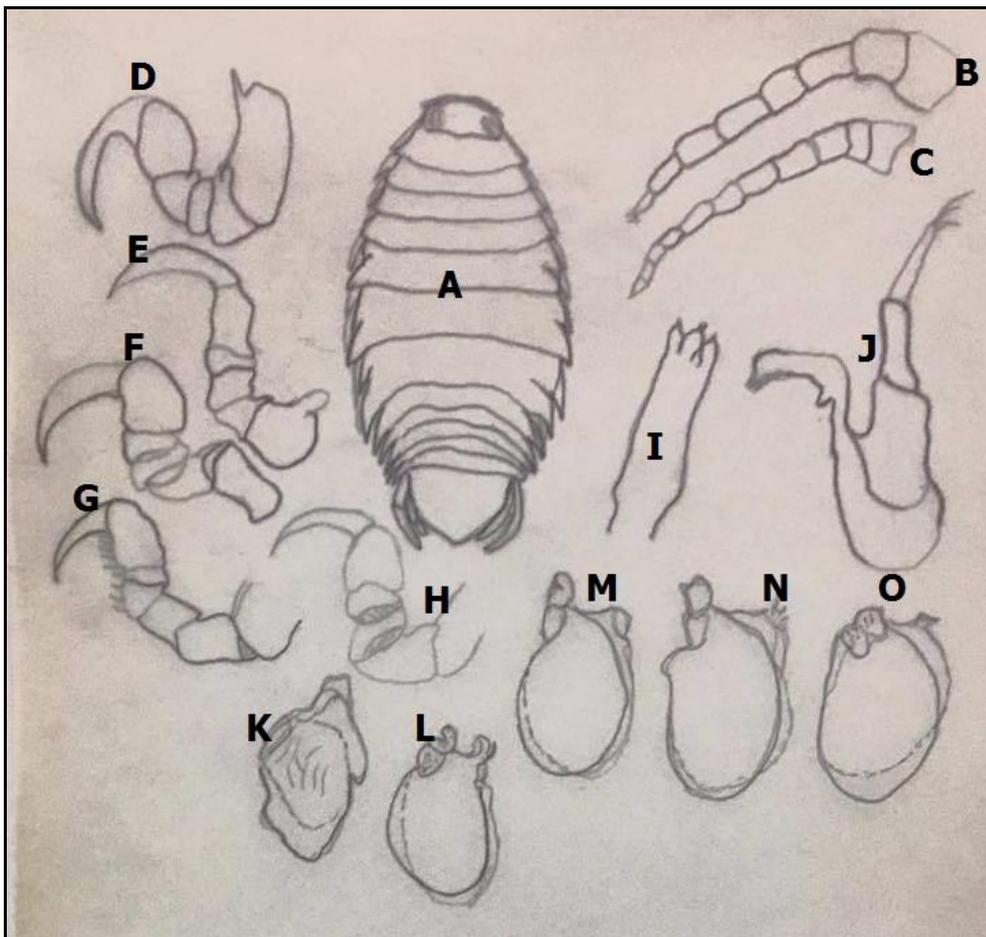
Female parasites were found attached to the skin of *S. vulgaris*.

**Description:** (based on 10 female specimens).

Female body size ranges from 15-25(20) mm in length and 8.-12 (10) mm as maximum width ( about 2.0 times as long as wide).



**Plate(2): A: Showing female *N. orbigny* attached to the base of pectoral fin of *S. vulgaris*. *Nerocila orbigny*: (B; Dorsal view), (C; ventral view) Scale bar =10mm**



**Figure 2: A, *N. orbigny* female dorsal view; B, 1<sup>st</sup> antenna; C, 2<sup>nd</sup> antenna ; D, pereopod1; E ,pereopod2; F, pereopod 3; H, pereopod 5; G, pereopod 7; I, Maxilla; J, mandible; M, pleopod 1; N, pleopod 2; K, pleopod 3; O, pleopod 4 ; L, pleopod 5.**

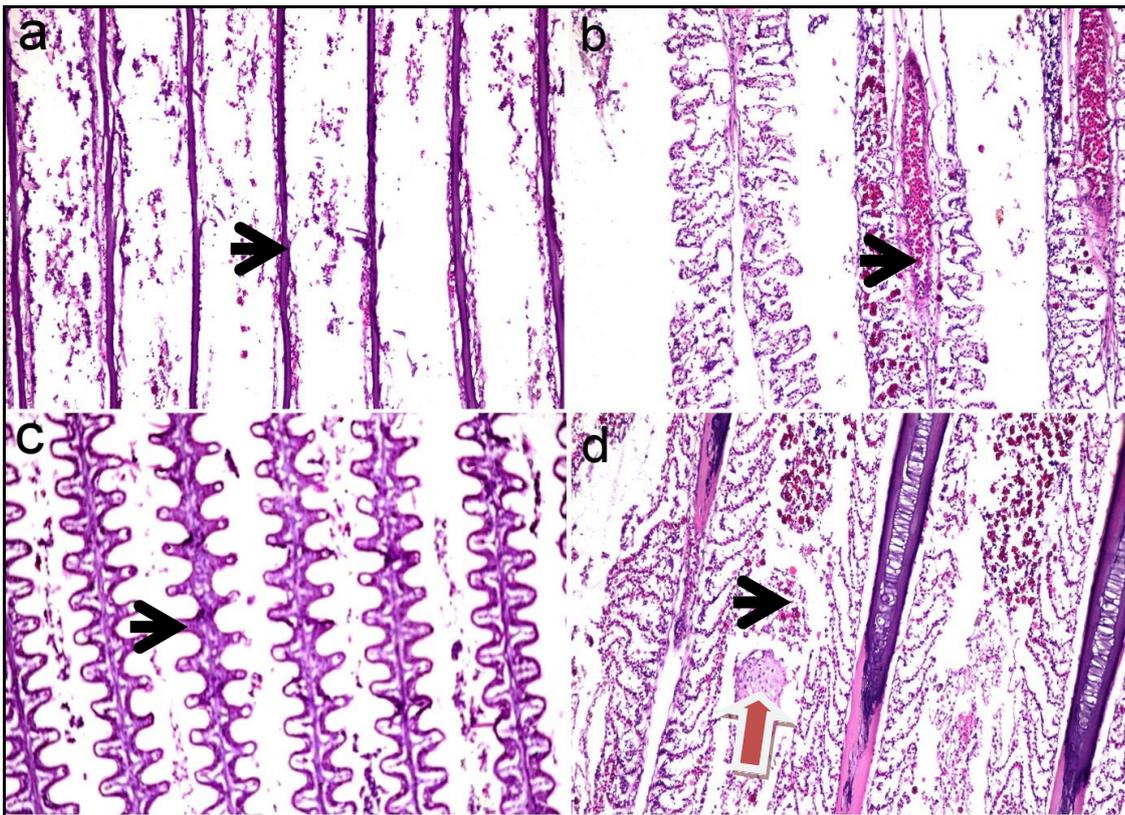
**Cephalon:** Anterior margin has an indistinct medial point. Eyes are small. Articles I and II of antennule are partly fused; antenna consists of 11 articles. Article 1 of Mandible palp is the longest while article 2 and 3 are subequal in length; article 3 with about 4 small and 6 long setae on the distolateral margin. Article 3 of maxilliped have 4 recurved spines.

**Pereon:** Pereonites I-IV posterolateral angles are not produced while pereonites V-VII posterolateral angles are produced and acute. The posterior margins of coxae of pereonites II-IV are rounded and coxae of pereonites V-VII are with acute posterior margins.

**Pleon:** Pleonite I is the longest. The ventrolateral margins of pleonites I and II are posteriorly directed and extend to pleonite V. Lateral margins of pleonites III-V are weakly produced. Pleotelson lateral margins are angled then converging to a caudomedial point. Pereopods 6 and 7 are subequal in size, each with carina on mediolateral margin. Uropod exopod is curving medially and is about 1.5 times as long as endopod. Endopod is straight and its distal margin is obliquely truncate.

### 3.3.Histopathological examination:

The histopathological examination of The gill lamellae on which the parasite was settled, showed complete sloughing of the epithelium lining the secondary gill lamellae (Plate 3a). The primary gill lamellae exhibited severe congestion with edema in secondary gill lamellae (Plate 3b). Hyperplasia of lamellar epithelium and mucous secreting cells with a fusion of secondary gill lamellae were also noticed (Plate 3c). Furthermore, the gill filaments were infiltrated by mononuclear cells and eosinophilic granular cells (EGC) with inflammatory exudates and a longitudinal section of the female isopod body (marsupium containing eggs) found between the gill filaments (Plate 3d).



**Plate 3:** Gills of fish infected with isopod (a) Gills showed complete necrosis and sloughing of the secondary gill lamellae (X100). (b) The secondary gill lamellae had edema and inflammatory cells infiltrate (X200). (c) Hyperplasia of lamellar epithelium and mucous secreting cells in the secondary gill lamellae (X100). (d) Presence of inflammatory exudates between the gill filaments and a longitudinal section of the female isopod body showing the marsupium containing eggs (red arrow) (X200). H & E stain.

#### 4. Discussion

Isopods recorded to parasitise many species of commercially important fishes worldwide causing significant economic losses to fisheries<sup>19</sup> where the incidence and intensity of parasitic isopods exhibit a considerable variation<sup>20,21</sup>.

The present study investigated the problem of isopod infestation among Lake Qarun fishes during the period from March to August 2015 through parasitological and pathological studies. Results revealed total prevalence of 32.66% with isopod infestation among *D. Labrax*, *S. vulgaris* and *T. zilli* fish species, the rate which is considerably higher than that recorded in Egypt for isopod infestation by<sup>22</sup> 8.62%,<sup>11</sup> 9% and<sup>12</sup> 10.26% from Mediterranean sea at Port-Said, Matroh, and Alexandria provinces respectively. Also it is higher than reported by<sup>23</sup> 19% among Mediterranean Sea fishes during the summer season. Also from Suez Canal in Ismailia province<sup>24</sup> 4%. These prevalence variations might be attributed to the differences in the periods of investigation, the examined fish species and the geographical distribution of fish hosts and parasites.

Regarding the detected isopod species, *R. thresherorum* adult female was isolated from the gills of *D. labrax* (42%) and *T. zilli* (12%) also small juveniles of this species were detected in the buccal cavity of three *T. zilli* fish. The isolated *R. thresherorum* in the present study is morphologically agreed with that reported by<sup>24</sup> from dorsal and pectoral fins and buccal cavity of the *D. labrax* and also with that reported by<sup>25</sup> from gills of the same host but with lower rates (6% and 25% respectively). According to the available literature, no previous records of the isopod *R. thresherorum* from Qarun lake, so the present investigation provides the first record of this isopod spp. from the gills of *D. labrax* and as juveniles from the buccal cavity of *Tilapia zilli* in a new locality.

In the present work, *N. orbignyi* was isolated from the skin of *S. vulgaris*. Morphologically, this detected isopod species is in agreement with that described by<sup>26</sup>. As the first record in Yemen from two Red Sea mugilid species (*Moolgarda sehili* and *liza aurata*) with an infestation rate of 4.2% and 7.7% respectively. Also, it agrees with that reported by<sup>27</sup> who isolated the same species from cultured *D. labrax* but with a lower rate of 6%. *Nerocila orbignyi* was also isolated from the gill chambers of *Tilapia zilli* collected from Qarun lake by<sup>28,29</sup>.

As the pathological impacts on fish skin due to *Nerocila orbignyi* infestation were previously investigated by several authors<sup>24,25,29</sup>, the present study estimated the histopathological alterations observed in the fish gills infested with *R. thresherorum*. In the current study, the histopathological alterations in the infested gills were similar to the findings reported by<sup>30,31,24,29</sup>. The detected deleterious impacts such as complete necrosis and sloughing of the secondary lamellae might be attributed to the pressure of the parasite feeding and its direct contact with the large sized isopod species<sup>32</sup>.

#### 5. Conclusion

Results of the present study cleared that, fish losses and marked drop in fish production from Lake Qarun could be regarded to the high prevalence of parasitic isopods causes severe impacts on the infected fish hosts. Further investigations are needed for estimating the problem of isopod invasion among Lake Qarun fishes.

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