Egyptian Journal of Aquatic Biology & Fisheries Zoology Department, Faculty of Science, Ain Shams University, Cairo, Egypt. ISSN 1110 – 6131 Vol. 27(4): 597 – 613 (2023) www.ejabf.journals.ekb.eg



# Distribution of some parasitic copepods on Actinopterygii fish from Egyptian water of Alexandria coast, with referencing to the histopathological alteration

Osama E. Zayed<sup>a</sup>, Ahmed M. Hellal<sup>a</sup>, Amr F. Zeina<sup>a</sup>, Safaa I. Tayel<sup>b</sup>

<sup>a</sup> Zoology Department, Faculty of Science, Al-Azhar University, Cairo, Egypt

<sup>b</sup> National Institute of Oceanography and Fisheries (NIOF), Egypt

\*Corresponding author: <u>osama\_sayed90@azhar.edu.eg</u>

# ARTICLE INFO

Article History: Received: July 7, 2023 Accepted: Aug.2, 2023 Online: Aug. 10, 2023

#### Keywords:

Parasitic copepods, Mediterranean Sea, Histopathology, Marine fish

#### ABSTRACT

During the year 2021, 870 Actinopterygii fish belonging to four different species were gathered along the Alexandria coast and examined for parasitic copepods. Four different species of parasitic copepods were reported during the period study including Lernanthropus kroyeri Beneden, 1851 from Dicentrarchus labrax (Linnaeus, 1758); Caligus affinis Heller, 1866 reported from Pomatomus saltatrix (Linnaeus, 1766); Mitrapus oblongus Pillai, 1964 from Sardina pilchardus (Walbaum, 1792); and Naobranchia cygniformis Hesse, 1863 were discovered from Boops boops (Linnaeus, 1758). Two of these four species (C. affinis and N. cygniformis) were reported from the Alexandria coast for the first time. Additionally, we recorded a few new host records, one of which included the discovery of M. oblongus on S. pilchardus. Parasitological indices known as prevalence, host-specificity of collected parasites, and their main micro-niches on investigated host species are done, as well as histopathological alternation, was discussed. The inclusion of new records of some parasitic species (C. affinis and N. cygniformis) in the Egyptian ecosystem contributes to enhancing their geographical distribution.

#### **INTRODUCTION**

The entire around, parasitic copepods are renowned for their variety and exceptional parasitism of commercially significant food fish species (Pillai, 1985; Johnson *et al.*, 2004; Cunningham, 2006; El- Rashidi & Boxshall, 2010). Numerous disease outbreaks in marine aquaculture are brought on by parasitic copepods (Maran *et al.*, 2009), without paying particular attention to the parasitic copepods, marine fauna predominated most of the studies on the fauna of Mediterranean copepods hundreds of years ago. More than 2000 different species of copepod parasitize freshwater and marine fish, and the majority of them are ectoparasitic, meaning they live in protected microhabitats that are always in contact with the outside world, such as the external nares, the eyes, the oral and branchial cavities, the gills, and the cloaca (Rosim *et al.*, 2013). Globally and along the Egyptian coast, where they have infested commercially important Actinopterygii fish species, copepod parasites have been the subject of intensive research.

Parasitic copepods species observed here belonging to order the Siphonostomatoida which now includes the families Lernaeopodidae Milne Edwards, 1840, and Lernanthropidae



#### Zayed et al., 2023

Kabata, 1979 (Khodami et al., 2017). Members of the genera Lernanthropus, Mitrapus, Caligus, and Naobranchia belong to the families Lernanthropidae Kabata, 1979, Caligidae (Burmeister, 1835), and Lernaeopodidae (Milne Edwards, 1840), respectively, are among the most widespread genera of parasitic copepods in marine environments worldwide (Kabata, 1979), except for the genus Naobranchia it has a limited spread (Madinabeitia & Nagasawa 2011). Lernanthropus sp. attaches to the gill filaments with its second and third sets of legs and causes the host to experience severe pathogenic responses that might lead to host loss and a significant drop in fish populations (Korun & Tepecik, 2005). Lernanthropus kroyeri has been discovered in many locales along the northern coast of Africa, the Adriatic Sea, and the southern North Sea in Europe (Kabata, 1979). Furthermore, L. koryeri is found across the Egyptian marine environment, ranging from the Red Sea to the Mediterranean Sea via the Suez Canal. Mitrapus oblongus was first discovered by El-Rashidy & Boxshall (2009) on immigrant and native clupeid fish captured in Egyptian coastal waters off Alexandria on Sardinella aurita. Caligus species are harmful pests that pose significant economic issues in commercial aquaculture and marine fisheries (Johnson et al., 2004). Caligus affinis was observed on three hosts in the Mediterranean Sea, Alexandria. One is Pomatomus saltatrix (Linnaeus, 1766), according to **Boxshall & El-rashidy** (2009). Naobranchia is a taxonomically diverse genus that belongs to the order Siphonostomatoida and is considered a gills-specific parasite for marine fishes. The genus Naobranchia has 23 species, (Dippenaar & Jordaan 2008; Madinabeitia & Nagasawa 2011). According to Tansel & Fatih (2012), parasitic copepod infections are becoming increasingly widespread in marine environments and aquaculture and have been associated with significant economic losses in the Mediterranean area due to morbidity and mortality in farmed marine fish (Papoutsoglou et al., 1996; Oktener & Trills, 2009). In Egypt, lesions were detected on the head area, the buccal cavity, the palate, the tongue, and the base of the gill arch of Dicentrarchus labrax and the skin of *Mugil cephalus*. These parasites were also discovered in seabass, mullet buccal, and branchial cavities (Ragias et al., 2004).

Parasitic copepods cause direct harm to their hosts through attachment mechanisms and feeding activities. All parasites are host-specific, albeit the level of host restriction varies greatly. Even parasite species with a diverse host range may prefer one host over another. As a result, **Rohde (1984)** distinguished between host range and host specificity. Host range refers to the total number of host species that a specific parasite may infect, regardless of how frequently or strongly the various host species are affected. The consequences and problems that parasitic copepods have on fish in the Mediterranean environment generally will be clearly illustrated with the aid of a few well-known species. Beyond this simple estimation, we sought to clarify some biological features, such as which species are preferred hosts, and to determine whether environmental changes could be shown to affect the species richness or distribution of parasitic copepods among the host fish. The infection's frequency and/or severity are factors in host specificity.

## **MATERIALS AND METHODS**

#### i. Study areas and Samples collection

This study was conducted during the year 2021 along the Alexandria coast of the Mediterranean Sea between 31°22'36" N, 30°01'39.1" E, from Abu-Qir Bay, and 31°12'23.4" N, 29°47'36" E, at Al-Anfoushi region. A total of 870 fish specimens belonging to four species of Actinopterygii fish were gathered and examined for parasitic isopods. All collected fish specimens were transported to the Invertebrate Microscopy Lab at the Zoology Department, Faculty of Science, Al-Azhar University for further investigations. For each fish, the total and the standard length, weight were measured, as well as fish's sex was determined. Fish taxonomy and host nomenclature were carried out according to Fish-Base (**Froese & Pauly 2018**).

#### ii. Histopathological examination

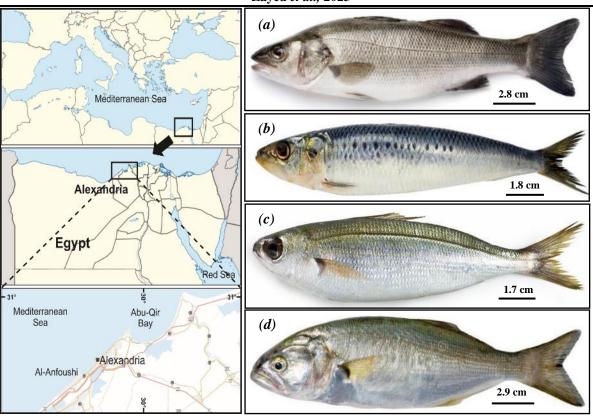
Immediately After dissecting the fish, gills were carefully removed, and small pieces were fixed in 10% formalin at 40 C, dehydrated in ascending grades of alcohol, and cleared in xylene. The fixed tissues were embedded in paraffin wax and sectioned into 5 micrometers in thickness. Sections were stained according to Harris Hematoxylin and Eosin method (**Bernet** *et al.*, **1999 as cited by Ahmed** *et al.*, **2022**). These sections were examined microscopically and photographed using a microscopic camera.

#### iii. Host specificity

To gauge the host specificity index, the host specificity index and average intensity of infection are as follows, according to **Rohde (1994)**:

$$Si \text{ (intensity)} = \frac{\sum \left(\frac{xij}{nijhij}\right)}{\sum \left(\frac{xij}{nij}\right)}$$

Where Si = host specificity of *i*th parasite species, xij = the number of parasites individuals of *i*th species in *j*th host species, nj = the number of host individuals in *j*th species examined, hij = rank of host species *j* based on the intensity of infection xij/nj (intensity of infection). Numerical values for the indices vary between 0 and 1, the higher the degree of host specificity, the closer to 1.



**Fig. 1.** Collection sites from the Mediterranean Sea coast of Alexandria, Egypt

Fig. 2. Infested hosts by various parasitic crustaceans,
a) Dicentrarchus labrax; b) Sardina pilchardus; c)
Boobs boops; and d) Pomatomus saltatrix.

#### RESULTS

During the period of study, four different fish species were collected and examined for parasitic copepods. Four different species of parasitic copepods (Fig. 3) were gathered host species including *Lernanthropus koryri* Beneden, 1851 from *Dicentrichus labrex*, *Caligus affinis* Heller, 1866 from *Pomatomus saltartix*, *Mitrapus oblongus* Pillai, 1964 from *Sardina pilchardus*, and *Naobranchia cygniformis* Hesse, 1863 from *Boops boops*.

## **Environmental factors**

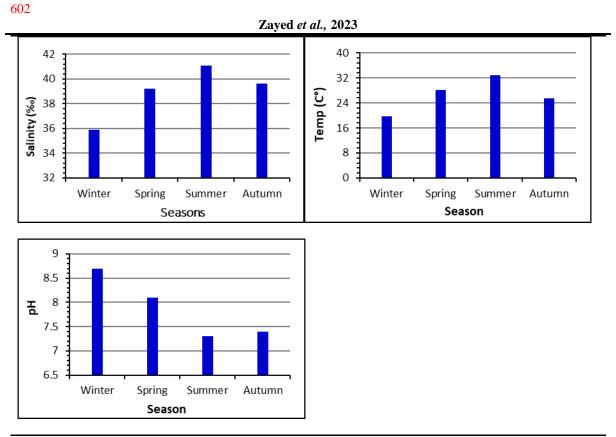
The results of the current study's analysis (Table 1 and Fig. 4) showed that the winter season had the lowest recorded salinity at 34.9 0.49 and the summer season had the greatest salinity at 41.4 0.65 and a temperature of 32.8 0.77 °C. However, the lowest hydrogen ion concentration was 7.3 0.16 in the summer and the highest value was 8.7 0.12 in the winter. In terms of salinity, the summer and winter seasons differed considerably (P<0.05) from the autumn and spring seasons, although the autumn season did not differ significantly (P>0.05) from the spring. All seasons show a significant change in recorded temperature (P<0.05), and between the winter and spring seasons as opposed to the autumn and summer seasons, there was a significant change in hydrogen ion concentration (P<0.05). When compared to the summer, the fall season did not differ substantially (P>0.05) (Table, 1). The aforementioned data leads us to the conclusion that summertime has the greatest salinity and temperature levels. and the lowest pH values.



**Fig. 3.** Parasitic copepod species. A, B) *Mitrapus oblongus*, (Pillai, 1964); C, D) *Lernanthropus kroyeri*, Beneden, 1851; E) *Caligus affinis*, Heller, 1866; F – K) *Naobranchia cygniformis*, Hesse, 1863, adult 9, for different forms. Scalebar=1mm.

Seasons	Salinity (‰)	Temperature (C°)	pН	
Winter	$35.9\pm0.49$	$19.6\pm0.29$	$8.7\pm0.12$	
Spring	$39.2\pm0.24$	$28.2\pm0.32$	$8.1\pm0.21$	
Summer	$41.1\pm0.65$	$32.8\pm0.77$	$7.3\pm0.16$	
Autumn	$39.6\pm0.36$	$25.4\pm0.30$	$7.4\pm0.16$	

 Table 1. Seasonal variations of environmental factors.



**Fig. 4.** Charts representing the average recorded value for Salinity, Temperature, and Hydrogen ion concentration during the study period.

## Parasitological indices of the collected parasitic copepods

Analysis of the current study (Table 2 and Fig. 5) showed that 370 specimens of parasite copepods were collected from four distinct species, representing 116 hosts, after evaluating 2730 fish specimens from nineteen different species.

# 1. Lernanthropus kroyeri Beneden, 1851

*Lernanthropus kroyeri* parasite was gathered from the *Dicentrarchus labrax*. These parasitic species exhibit a higher prevalence on their hosts in comparison to other parasite species during the study period (P=29.1%). In terms of host species versus seasons, the highest prevalence value was reported during the summer season (P=39.7% and MI=3.7 per host), while the lowest prevalence rate was recorded during the winter season (P=24.1% and MI=3.8).

# 2. Naobranchia cygniformis Hesse, 1863

*Naobranchia cygniformis* parasitized *Boops boops* gills, with an annual prevalence (P=21% and MI=2.1). In terms of host species versus seasons, the highest prevalence was recorded during the summer season (P=34.1% and MI=2), but the lowest prevalence was reported during the spring season (P=10% and MI=1.8).

# 3. Caligus affinis Heller, 1866

Caligus affinis was found on the buccal cavity and gill regions of Pomatomus saltatrix only, with an annual prevalence rate (P=8.1% and MI=3.2), In terms of host species versus seasons, the highest prevalence rate was reported during the winter season

(P=11.1% and MI=4), but the lowest prevalence rate was reported during the autumn season (P=5.9% and MI=3.2).

## 4. Mitrapus oblongus Pillai, 1964

*Mitrapus oblongus* was recorded on the gills of *Sardina pilchardus*, with an annual prevalence rate (of P= 2.7% and MI= 2.5), In terms of host species versus seasons, the highest prevalence rate was reported during the winter season (P=4.1% and MI= 3.6), but the lowest prevalence rate was reported during the autumn season (P=1.2% and MI=3.0).

Analysis of variance for *L. Kroyeri* (Fig. 5) revealed a substantial variation (P< 0.05) for host species, but no significant differential (P> 0.05) across various seasons, and R<sup>2</sup> is valued at 91.43%. Data analysis of *C. affinis* observed a significant variance (P< 0.05) for host species. Similarly, there is no significant difference (P> 0.05) over seasons and R<sup>2</sup> is valued at 68.29%. An analysis of Variance for *M. oblongus* shows a significant variance (P< 0.05) for host species (*S. pilchardus*), while there was no significant differentiation (P> 0.05) over seasons and R<sup>2</sup> valued at 70.40%. The analysis of variance for *N. cygniformis* reveals a significant variation (P<0.05) for host species but no significant distinctions (P> 0.05) across seasons, with R<sup>2</sup> valued at 86.17% as shown in Figures (4*a* & *b*). The results concluded that all parasitic species prevail in the summer season in comparison to other seasons, except *C. affinis* prevails in the spring season.

## Relation between the environmental factors and prevalence of parasitic copepods

Under the number of parasites collected from its host species throughout the research period, the abundance and distribution of parasitic copepods were calculated. According to the data, temperature, pH, and salinity, all correlated positively with the abundance of the parasite species *Lernanthropus kroyeri*, *Caligus affinis*, *Mitrapus oblongus*, and *Naobranchia cygniformis* that were being studied, except for *C. affinis*, which had a mildly negative connection with salinity. Also, there was a weakly negative correlation in pH with *L. kroyeri* and *N. cygniformis*. This indicates that a built link exists between the predominance of parasitic copepod species and high salinity and temperature levels. Although there is only a weakly documented correlation with the other parasite species that have been observed, *L. kroyeri* and *N. cygniformis* were significantly correlated with physical factors, showing a strong positive connection with salinity and temperature and a strong negative connection with pH.

## Micro-niche, Host specificity

Only the gills of *Dicentrarchus labrax* were micro-niche to get *Lernanthropus kroyeri*, 250 parasite specimens were plucked from the gills of 68 fish hosts constituting 100% parasitism. Additionally, *Caligus affinis* was isolated from the mouth and gills of their hosts, where 42 parasite organisms were cut from 13 hosts, with (76.9%) coming from the mouth, and 10 parasite organisms on the gills (23.1%). *Mitrapus oblongus* was recovered solely from the host species *Sardina pilchardus*, all infection locations on host gills were documented, and 25 parasite organisms were cut from 10 hosts, representing 100%. Only the

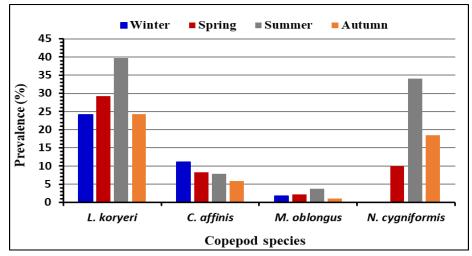
Zayed et al., 2023

*Boobs boops* host species was used to get *Naobranchia cygniformis*, and all infection sites on the host gills were noted and clipped. 53 parasite individuals were discovered from 25 hosts, as shown in Table (3) and Figs. (6 & 7).

Copepod species	Host species	Seasons	NEF	NIF	Prevalence		MI
					P. No	(%)	1711
Lernanthropus koryeri	Dicentrarchus labrax	Winter	54	13	50	24.1	3.8
		Spring	48	14	51	29.2	3.6
		Summer	58	23	88	39.7	3.8
		Autumn	74	18	61	24.3	3.9
		Total	234	68	250	29.1	3.8
Caligus affinis	Pomatomus saltatrix	Winter	27	3	12	11.1	4.0
		Spring	48	4	13	8.3	3.3
		Summer	51	4	12	7.8	3.0
		Autumn	34	2	5	5.9	2.5
		Total	160	13	42	8.1	3.2
Mitrapus oblongus	Sardina pilchardus	Winter	73	3	5	4.1	3.6
		Spring	94	2	10	2.1	5
		Summer	109	4	7	3.7	1.8
		Autumn	81	1	3	1.2	3
		Total	357	10	25	2.7	2.5
Naobranchia cygniformis	Boops boops	Winter	-	-	-	-	-
		Spring	40	4	7	10.0	1.8
		Summer	41	14	28	34.1	2.0
		Autumn	38	7	18	18.4	2.6
		Total	119	25	53	21.0	2.1

Table 2. List of distribution seasonally of parasitic copepod species among their host species.

*NEF*= *No. of Examined fish; NIF*= *No. of Infested fish; MI*= *Mean Intensity, and P*= *parasitic Number.* 



**Fig. 5.** The chart represents the prevalence of parasitic species at different seasons concerning host species.

According to ANOVA analysis, as for the sites of infection, results indicated that the gills site at all infested fish species varied significantly (P> 0.05) compared to other sites, either the mouth or buccal cavity. The results suggested that there was a significant variance (P< 0.05) in gills infection for *D. labrex* in comparison to other infested fish species when compared to recorded count infection of gills (P> 0.05). For the sites of infection vs seasons, the results revealed a significant differentiation (P< 0.05) for the summer season with the gills site of infection in comparison to other seasons at different infested fish species at different infection sites, except for Autumn (P> 0.05).

Host specificity refers to the number of host species infected by a parasite species, irrespective of how frequently or heavily the various species are infected. The results suggested host specificity for all parasites (restricted to certain host species). Where the values *Si* ranged between 0 - 1, whereas the values gradually decreased from 0.31 in *C. affinis* to 0.27 in *L. koryri*, the higher values of host specificity approaching 1; however, even those species that have a wide host range may prefer some hosts over others. Knowing host specificity considers the prevalence and/or intensity of infection. So, the current observation revealed the host specificity of parasite organisms.

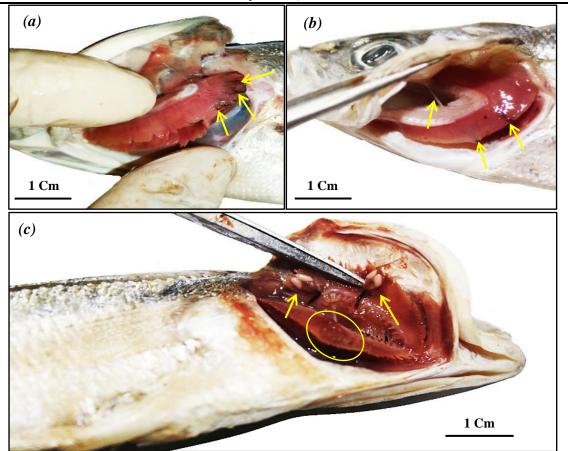
A clinical evaluation of fish with various parasite copepod species found an infestation that was more severe than that of the majority of the host species. The clinical investigation revealed the following. Increased opercular movement, gasping for air at the water's surface, and clustering near an air source are the first two signs of increased breathing frequency. According to post-mortem lesions, the gills were engorged, hyperemic, and covered in mucous. Additionally, the gills were damaged, hemorrhagic, and inflamed as a result of the parasites' feeding behaviors (feeding on the epithelium and mucous), which resulted in an excessive amount of mucous secretion as shown in (Figs. 6 & 7).

Parasite species	Host species	Р.	H.	Micro-niche		Si
		No.	No.	Gills	BCM	- 51
L. koryeri	D. labrax	250	68	250	-	0.27
C. affinis	P. saltatrix	42	13	10	32	0.31
M. oblongus	S. pilchardus	25	10	25	-	0.40
N. cygniformis	B. boops	53	25	53	-	0.48

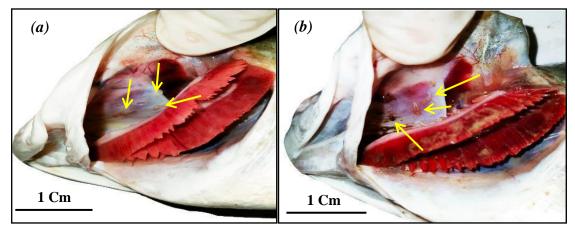
Table 3. Host specificity and the number of parasitic copepods species on various fish host

BCM = Buccal cavity and mouth. P = parasitic number H = Host number Si = Host specificity

Zayed et al., 2023



**Fig. 6.** a) Infested of *D. labrex* by *L. kroyeri*; b) shows the gills were congested and hyperemic and covered with much mucous and c) Showing infested of *B. boops* gills by *N. cygniformis* and show gill ulcer and erosion at the site of parasite attachment.

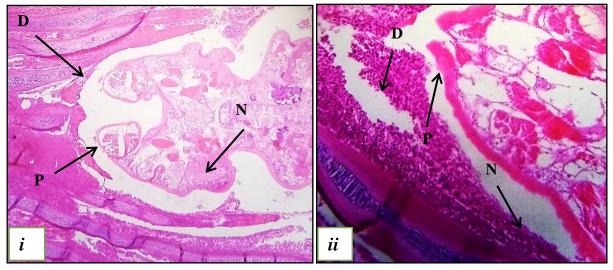


**Fig. 7.** a) Infested *Pomatomus saltatrix* in the buccal cavity and gills by *Caligus affinis;* b) shows damaged, hemorrhagic, and inflamed due to parasite-feeding behavior with excessive mucous secretion.

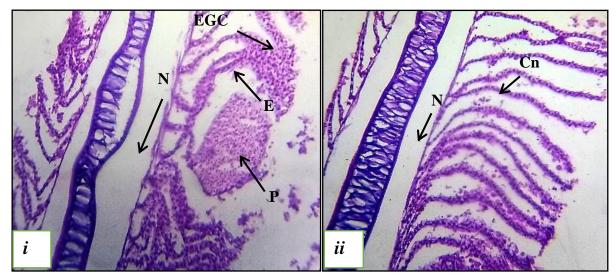
# Histopathological examination of examined fish

Examining infested gills hosts with pre-adult and adult stages of *L. kroyri* and *M. oblongus* species revealed parasitic (P) forms appeared in secondary gill lamellae, causing degeneration (D) of their epithelial cells at the site of parasitic attachment, leading to the formation of

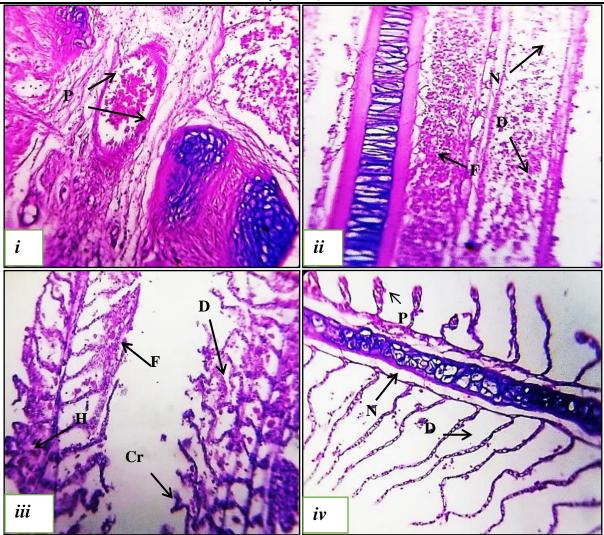
necrosis (N) focal area. In addition to epithelial cell hyperplasia (H), filament fusion (F) occurs. Curling (Cr) of secondary lamellae was also observed in several samples (Figs. 9 & 10i - ii). In rare circumstances, *L. Kroyri* with distinctive long egg sacs occurs between the gill filaments of *D. labrex* fish, causing epithelial cell degeneration and the creation of the necrotic (N) region around the parasite (P) (Figs. 8*i* and *ii*). Gills histopathological alterations of infested *B. boops* fish with the adult stage of *N. cygniformis* (P) species indicated that the infestation is usually accompanied by eosinophilic granular cells (EGC) aggregation with lymphocytes and edema (E) in the gill filaments together with congestion (Cn) of the branchial blood vessels. In addition to necrosis (N) in primary lamellae (Figs. 9*i* and *ii*).



**Fig. (8,** *i* **and** *ii***).** The Sagittal section of the gills of *D. labrex* is infested with *L. Kroyri*. Showed parasite (P) and degeneration (D) necrosis (N) in gill lamellae (H & E stain, X 200 and 400 respectively).



**Fig. (9** *i* **and** *ii*). The sagittal section of the gills of *B. boops* infested with *N. cygniformis* showed *parasites* (P), eosinophilic granular cells (EGC) aggregation with lymphocytes, necrosis (N), edema (E), and congestion (Cn) in gill filaments lamellae (H & E stain, X 400).



**Fig.** (10, i - iv). The sagittal section of the gills of *D. labrex* and *S. pilchardus is* infested with parasitic copepods. Showed *i*): section of the parasite (P) form between the secondary gill lamellae, *ii*): Marked degeneration (D), necrosis (N), and fusion (F) of secondary gill lamellae, *iii*): degeneration (D) hyperplasia (H), curling (Cr) and fusion (F) of secondary gill lamellae *iv*): Destruction (De) in epithelial cells of secondary lamellae at the site of parasitic (P) attachment and necrosis (N) in primary lamellae (H and E stain, X 400).

## DISCUSSION

The present study aimed to investigate parasitic copepod invasion on four commercial fish (*Dicentrichus labrax, Sardina pilchardus, Pomatomus saltratrix,* and *Boobs boops*) at the Eastern Mediterranean Sea of Alexandria coast. It seems that *Lernanthropus kroyeri* hosts only *D. labrax* in the Mediterranean Sea, the previous perception being consistent with the current study in host specificity in the fact that the seabass, *D. labrax,* is the only host of *L. kroyeri.* Approximately 1.5% daily mortality was recorded in sea bass *D. laprax* raised in a floating cage farm on the Aegean coast of Turkey; 15 seabass were examined for parasitological and pathogenicity (**Yardimci & Pekmezci, 2012**) outbreaks and deaths were observed caused by *L. kroyeri* is frequently found in seabass farming, and economic losses

occur as a result of reduced feed conversion, reduced growth, mortality, loss of product value and processing costs (Manera & Dezfuli 2003). In the present study, the prevalence of L. kroyeri was 29.1%, and the mean intensity of infection was 3.7 in the investigated sea bass; this result was lower than what has been obtained by Yardimci & Pekmezci, (2012), who recorded a prevalence of 100% and mean intensity 11.7; also, Toksen (2007) found prevalence as 100% and mean intensity of infection as 50.3. On the other hand, near results from our findings have been reported by Manera & Dezfuli (2003), who reported a prevalence of 35% and a mean intensity of infection of 10.86 in their study. Histologic examinations indicated L. kroyeri was present on branchial lamellae in the gills of all fishes. Lamellary edema formation, a fusion of the secondary lamellae, and necrosis in both primary and secondary lamellae due to parasitic irritation were determined that serious lesions and necrosis of the primary and secondary lamellae of the gills are a consequence of parasitism of L. kroyeri which causes more severe destruction, and clinical signs (Yardimci & Pekmezci, 2012). Three parasitic copepods new to the Mediterranean fauna were reported in waters off the Egyptian coast near Alexandria by El-Rashidy & Boxshall (2009); one of them, Mitrapus oblongus, is of Indo-Pacific origin and is considered to have invaded the Mediterranean through the Suez Canal on Red Sea immigrant hosts (El-Rashidy & Boxshall 2009). Here we address the second record of *M. oblongus* within our regional waters, while previous research did not address the possible negative effects of histopathological alternation due to the invasion of *M. oblongus* on *S. pilchardus* species. The present study aimed to refer to some ecological, biological, and histopathological changes and prevalence rates resulting due to invasion or parasitism on S. pilchardus species, the results revealed that there was a histopathological alternation caused by secondary gill lamellae destruction and epithelial cell hyperplasia with prevalence rate 2.7% among infested fish, and host specificity for this parasite was confirmed with a value of 0.40, In general, this brief investigation agrees with a set of articles published on the family Lernanthropidae which deals with the highest rates and seasons of infection and the impact of environmental factors on the prevalence of parasites (Ravichandran et al., 2009; Noor El-Deen, 2007).

The first record of *Caligus affinis* was conducted by **Brian (1935)** based on a single female collected from *Umbrina cirrosa* species at Genoa, then a second recorded on *Umbrina canariensis* at the Adriatic Sea; therefore, *P. saltratrix* is considered a new host species for *C. affinis* in the Egyptian Coasts within the Mediterranean Sea. The previous research did not address the effects caused by the invasion of *C. affinis* on their hosts, but here the consequences of parasitism on its host have been clarified by discussing infection rates, infection intensity, and histopathological changes in gills and skin infection caused by this parasitic species (**Woo, 2006; Noor El-Deen, et al., 2012 & 2013).** In general, *C. affinis* belongs to the family Caligidae, and previous research indicated that negative effect on Caligidae family hosts through histological changes, seasons, and rates of infection, which were revealed in some species within this family and the results we reached showed that our result came along to what was presented on previous research, it has been proven through the results that this species is the least in host specificity among its peers. *N. cygniformis* was recorded and described by **Hesse in 1863** parasitism on *Boops boops* and *Pagellus erythrinus* 

#### Zayed et al., 2023

caught in the Mediterranean Sea, and in 1877 (synonym *N. amplectus*) (Kurz 1877) from *Diplodus annularis* and *D. lineatus* caught in the Adriatic Sea. Since then, *N. cygniformis* has been reported several times from sparid fishes caught in the Mediterranean and Adriatic seas (**Raibaut** *et al.*, **1998; Madinabeitia & Nagasawa, 2011**). The present study suggested that there was an exacerbation of the disease as a result of infection, where the infection rates were recorded at 21%, the average infection intensity was 2.1, and the specificity of the host was 0.48, these results came near to what has been reported by **Baker** *et al.*, **(2005)** in the occurrence of *Naobranchia lizae* on *Mugil cephalus* the prevalence was 16.8% with mean intensity 2.79  $\pm$  0.26 and suggested that the occurrence rate was positively correlated with salinity. Also, the prevalence of *N. cygniformis* was previously reported only from *D. annularis*, at 19%, with a medium intensity (**Radujkovic & Raibaut 1989**), which is higher than the findings by **Dippenaar & Sebone**, **(2021)** of a prevalence of 3.9% and a mean intensity of 4 individuals per infected host which is lower than the findings in our study.

# Conclusions

The host's vulnerability to severe pathogenic reactions, which may result in host loss and a considerable drop in fish populations, is the main issue with parasitic copepod sampling. The results showed that parasitic copepods specialize in parasitizing specific species and are more attracted to some species than others, possibly because they are present in the same zonation. Our investigation focused on some patterns of distribution of parasitic copepods between their host species. The host specificity for parasitic species was determined. Aside from this straightforward estimate, unique procedures exclusive to histology have been used to track the dismal state brought on by this infiltration.

## Acknowledgments

This paper is based on work funded by the Science, Technology, and Innovation Funding Authority (STDF), under the Graduate Support Grant (PGSG) grant. Project No. 45018/2021, Academy of Scientific Research and Technology (ASRT), Egypt. We would also like to sincerely thank the anonymous referees for their helpful comments that enabled us to improve the quality of the manuscript.

## **Declaration of Competing Interest**

The authors declare that they have no known competing financial interests or personal relationships that could influence the work presented in this paper.

# **Conflict of interest**

I, as the corresponding author, declare, on behalf of all authors of the paper, that no financial conflict of interest exists concerning the work described.

## **Ethical approval**

The specimen is not under the listed category of experimental animals which needs ethical approval.

#### Data availability statement

Raw data was generated at the faculty of Science Al-Azhar University. Derived data supporting the findings of this study are available from the corresponding author on request.

## REFERENCES

- **Baker, T. G.; Pante, E. and Buron, I.** (2005). Co-occurrence of *Naobranchia lizae* (Copepoda) and *Metamicrocotyla macracantha* (Monogenea), gill parasites of the striped mullet *Mugil cephalus*. Parasitology Research 97: 515 520.
- **Bernet D.; Schmidt H.; Meier W.; Burkhardt-Holm P. and Wahli T.** (1999). Histopathology in fish: proposal for a protocol to assess aquatic pollution. J Fish Dis 22: 25–34.
- **Boxshall, G.A. and El- Rashidy, H.H.** (2009). A review of Caligus productus species group, with the description of a new synonymy and supplementary descriptions. Zoo taxa 2271: 1- 26.
- Brian, A. (1935). *Caligus parassiti* dei pesci del Mediterraneo (Copepodi). Annali del Museo Civico di Storia Naturale Giacomo Doria, 57: 152–211.
- **Cunningham, C.** (2006). A review of research and field data on relative louse infection levels on wild salmon smolts and sea trout and the proximity of fish farms to river estuaries fisheries research services, marine laboratory, 375 victoria road, aberdeen, ab11 9db.
- Dippenaar, S.M. and Jordaan, B.P. (2008). Description of the adult female and male of Naobranchia kabatana n. sp. (Copepoda: Lernaeopodidae) from Muraenesox bagio (Hamilton) (Muraenesocidae) caught in the Indian Ocean off South Africa, Syst Parasitol, 70:27–34.
- **Dippenaar, S.M. and Sebone, M.M.** (2021): The first report of *Naobranchia cygniformis* Hesse, 1863 (Copepoda: Lernaeopodidae) off South Africa, with a redescription of the adult female, African Journal of Marine Science, 43: 3, 413-416.
- **El-Rashidy H. and Boxshall G.A.** (2010). Parasitic copepods on immigrant and native clupeid fishes caught in Egyptian coastal waters off Alexandria.Syst Parasitol., 76(1): 19-38.
- El-Rashidy, H.H. and Boxshall, G. (2009). Parasites gained: aline parasites switching to native hosts. J. Parasitol., 95 (6): 1326-1329.
- Fischer, W.; Bauchot M. L. and Schneider, M. (1987). Fiches FAO d'identification des espèces pour les besoins de la pêche. (Révision 1). Méditerranée et Mer Noire. Zone de Pêche 37. Rome: FAO, 761–1529.
- Froese, R. and Pauly, D. (2018). Fish-Base. http://www.fishbase.org.
- Horton, T. Okamura, B. (2001). Cymothoid isopod parasites in aquaculture: a review and case study of a Turkish sea bass (*Dicentrarchus labrax*) and sea bream (*Sparus auratus*) farm. Diseases of Aquatic Organisms, 46: 181-188.
- Jithendran, K.P.; Natarajan, M. and Azad, I.S. (2008). Crustacean parasites and their management in brackish water finfish culture. Marine Finfish Aquaculture Network, July-September: 47-50.

- Johnson, S.C.; Treasurer J.W.; Bravo S.; Nagasawa K. and Kabata Z. (2004). A review of the impact of parasitic copepods on marine aquaculture. Zoological Studies, 43: 229-243.
- **Kabata, Z**. (1970). Crustacea as enemies of fishes. In Diseases of fish. (S.F. Snieszko and H.R. Axelrod, Eds.) pp. 1 171. TFH publications, Jersey City.
- **Kabata, Z.** (1979): Parasitic copepoda of Australian fishes, XII. Family Lernanthropidae, Crustaceana 37(2): 206-213.
- **Kabata, Z.** (1988). Copepoda and Branchiura. In Guide to the parasites of Fishes of Canada. Part II- Crustacea, Margolis, L. and Kabata, Z. (Eds.). Fish Aqua. Sci., 101: 3-127 pp.
- **Khodami, S.; McArthur J.V.; Blanco-Bercial, L. and Arbizu P.M.** (2017). Molecular phylogeny and revision of copepod orders (Crustacea: Copepoda). Scientific Reports 7: 9164.
- Korun, J. and Tepecik, R.E. (2005). Gill Lesions Caused by Infection of *Lernanthropus* spp. Blainville, 1822 on Cultured Sea Bass, *Dicentrarchus labrax*. Journal of Veterinary Faculty, Istanbul University 31: 1 8.
- Madinabeitia, I. and Nagasawa, K. (2011). Description of *Naobranchia denticis* n. sp. (Copepoda, Siphonostomatoida, Lernaeopodidae) parasitic on *Dentex hypselosomus* (Teleostei, Sparidae) from Japanese waters, with a key to the species of *Naobranchia*, *Crustaceana* 84: 461–476.
- Manera1, M. and Dezfuli, B.S. (2003). *Lernanthropus kroyeri* infections in farmed sea bass *Dicentrarchus labrax*: pathological features, Dis Aquat Org 57: 177–180.
- Maran, B.A.V.; Seng, L.T.; Ohtsuka, S. and Nagasawa, K. (2009). Records of Caligus (Crustacea: Copepoda: Caligidae) from Marine FishCultured in Floating Cages in Malaysia with a Redescription of the Maleof *Caligus longipedis* Bassett-Smith, 1898. Zoological Studies 48(6): 797-807.
- Noor El-Deen A.E.; Mona S.Z. and Shalaby, I.S. (2013). Some Investigations observed in Culture Seabass (*Dicentrarchus labrax* L.) infested with *Lernanthropus kroyeri* and *Nerocila orbignyi* and Exposed to Pollution during different seasons at Dammaitte province Life Science Journal. 10(3): 1877-1884.
- Noor El-Deen, A.E.; Abd El Hady, O.K. and Mona, S.Z.I. (2012). Field studies on *Caligus* disease among cultured *Mugil cephalus* in brackish water fish. Life Science Journal, Transport 9(3): 733-737.
- Noor El-Deen, A.I.E. (2007). Comparative studies on the prevailing parasitic diseases in mono sex tilapia and natural male tilapias in Kafr El Sheikh Governorate fish farms. Ph.D. Thesis, Fac. Vet. Med., Kafr El-Sheikh University, Egypt.
- **Oktener, A. and Trilles, J.P.** (2009). Four parasitic copepods on marine fish (Teleostei and Chondrichthyes) from Turkey. Acta Adriat. 50: 121-128.
- **Ozak, A.A.; Demirkale, I. and Yanar, A.** (2012). First record of two species of parasitic Copepods on immigrant Pufferfishes (Tetraodontiformes: Tetraodontidae) caught in the Eastern Mediterranean Sea. Turkish Journal of Fisheries & Aquatic Sciences, 12: 1–2.
- Papoutsoglou, S.; Costello, M.J.; Stamou, E. and Tziha, G. (1996). Environmental conditions at sea cages, and ectoparasites on farmed European sea bass *Dicentrarchus*

*labrax* and gilt-head sea bream *Sparus aurata* at two farms in Greece. Aquac. Res. 27: 25-34.

- **Pillai, N.K.** (1985). The fauna of India: Copepod parasites of marine fishes. Zoological Survey of India, Calcutta.
- Radujkovic, B.M. and Raibaut, A. (1989). Parasites des poisons marins du Monte ne gro: cope podes. Acta Adriatica, 30: 237–278.
- Ragias, V.; Tontis, D., and Athanassopoulou, F. (2004). Incidence of an intense Caligus minimus Otto 1821, C. pagetti Russel, 1925, C. mugilis Brian, 1935 and C. apodus Brian, 1924 infection in lagoon cultured sea bass (Dicentrarchus labrax L.) in Greece, 2004. Aquaculture 242: 727-733.
- Raibaut, A.; Combes, C. and Benoit, F. (1998). Analysis of the parasitic copepod species richness among Mediterranean fish. Journal of Marine Systems, 15: 185–206.
- Ravichandran, S.; Rameshkuniar, G. and Kumaravel, K. (2009). Variation in the morphological features of isopod fish parasites. World J. Fish Mar. Sci. 1 (2): 137–140.
- Rohde, K. (1984). Ecology of marine parasites. Helgola'nder Meeresuntersuchungen 37, 5-33.
- **Rohde, K.** (1994). Niche restriction in parasites: proximate and ultimate causes. Parasitology 109, 69-84.
- Romero, R.C. and Öktener, A. (2010). *Mitrapus oblongus* (Pillai, 1964) (Copepoda: Siphonostomatoida: Lernanthropidae) female and male redescription from specimens collected on *Sardinella aurita* from Turkey. Bull. Eur. Ass. Fish Pathol., 30(4). 120.
- **Rosim, D.F.; Boxshall G.A. and Ceccarelli P.S.** (2013). A novel microhabitat for parasitic copepods: A new genus of Ergasilidae (Copepoda: Cyclopoida) from the urinary bladder of a freshwater fish. Parasitol. Int. 62:3 47-354.
- Shaheen, A. (2013). An industry assessment of tilapia farming in Egypt. African Union Inter-African Bureau for Animal Resources (AU-IBAR).
- Tansel, T. and Fatih, T.P. (2012). Ectoparasitic sea lice, *Caligus minimus* (Otto 1821, Copepoda: Caligidae) on *Brawn wrasse*, *Labrus merula* L., in Izmir Bay, Aegean Sea, Italian Journal of Animal Science, volume 11: 38.
- **Toksen, E.** (2007). *Lernanthropus kroyeri* van Beneden, 1851 (Crustacea: Copepoda) infections of cultured sea bass (*Dicentrarchus labrax L.*). Bull Eur Ass Fish Pathol, 27: 49-53.
- Woo, P.T.K. (2006). Fish diseases and disorders. CAB, Int. Wallingford, Oxon, Uk., pp: 482-565.
- **Yamaguti, S.** (1936). Parasitic copepods from fishes of Japan. Part 2. Caligoida, I, 1–22, pls 1–12.
- **Yardimci, B. and Pekmezci, G.Z.** (2012). Gill histopathology in cultured sea bass (*Dicentrarchus labrax* (L.) coinfected by *Diplectanum aequans* (Wagener, 1857) and *Lernanthropus kroyeri* (van Beneden, 1851), Ankara Üniv Vet Fak Derg, 59: 61-64.