## **QATAR UNIVERSITY**

### COLLEGE OF ARTS AND SCIENCES

### DIVERSITY OF METAZOAN PARASITES OF TWO REEF ASSOCIATED

## FISH SPECIES FROM QATAR (ACANTHOPAGRUS BIFASCIATUS AND

POMACANTHUS MACULOSUS)

BY

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A Thesis Submitted to
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Sciences

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of the Requirements

for the Degree of

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ABSTRACT

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Title: Diversity of Metazoan parasites of two reef associated fish species from Qatar

(Acanthopagrus bifasciatus and Pomacanthus maculosus)

Supervisor of Thesis: Radhouan Ben-Hamadou.

The aim of the present work was to study the parasitic diversity of two coral-reef

associated fish species (Acanthopagrus bifasciatus and Pomacanthus maculosus) from

Qatari waters. There were eight species of parasites encountered during the present

study: five ecto-parasites (Polylabris mamaevi, Alella sp., Caligus haemulonis,

Lernanthropus sarbae and Pranzia larva of Gnathia africana) and three endo-parasites

(Stephanostomum sp., Dujardinascaris sp. and Cucullanus sp.). Description of the

encountered parasites and comparison with previously described related species was

conducted.

According to present data, four parasites were considered as new records in the

Arabian Gulf (Polylabris mamaevi, Caligus haemulonis, Lernanthropus sarbae and

Pranzia larva of *Gnathia Africana*) and seven parasites are considered as new records in

Qatari waters (Polylabris mamaevi, Alella sp., Caligus haemulonis, Lernanthropus

sarbae and Pranzia larva of Gnathia Africana, Dujardinascaris sp. and Cucullanus sp.)

Acanthopagrus bifasciatus is considered a new host for five parasites (Polylabris

mamaevi, Alella sp., Caligus haemulonis, Lernanthropus sarbae and Pranzia larva of

Gnathia Africana), while Pomacanthus maculosus is considered a new host for three

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parasites (*Polylabris mamaevi*, *Stephanostomum* sp. and Pranzia larva of *Gnathia africana*.).

Anatomical observations in the fish host, microhabitat of the parasites, mixed infection, host specificity, relationship between fish size and infection, spatial variations and parasite infection and gut contents are also discussed in detail.

# **DEDICATION**

To my family.

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#### CHAPTER1: INTRODUCTION AND LITRATURE REVIEW

Worldwide, especially in coastal cities the marine environment not only provides important food resources but also critical goods and services to human society. Fish and other marine products are also processed into pharmaceuticals, nutraceuticals and other valuable co-products. Fishing and diving are popular entertainments worldwide.

Globally more than 70 % of the earth is covered with water and nearly 30% of people suffer from malnutrition, hence come the importance of fish as major component of the global food basket to enhance the nutrition, health, and wellbeing of humans (Tacon & Metian, 2013; Thilsted *et al.*, 2014).

The global fish production in 2014 is estimated to be 167.2 Million Tonnes of fish (93.4 Million Tonnes total capture and 73.8 total aquaculture) while per capita food fish supply is 20.1 kg (FAO, 2016).

Health assessment of marine systems using biological indicators is considered an important tool for water resources management (Diamond, 2003; Simon, 2003; Borja *et al.*, 2008). Namely, fishes are used as environmental-integrity indicators of aquatic ecosystems (Whitfield & Harrison, 2008; Guillemot *et al.*, 2014), due to their differential life strategy and vulnerability to environmental conditions (Scharbert & Borcherding, 2013).

Accordingly, examination of fish health can reflect the ecological quality status of the receiving marine environment and the range of pressures on the studied ecosystems (Pérez-Domínguez *et al.*, 2012). Fish health status is recognized as a valuable indicator of environmental integrity status (Borja *et al.*, 2011) and potentially applicable to the highly impacted Qatar marine environment.

Fish parasites can be divided according to their habitats into two groups:

endoparasites which live inside the host and ectoparasites which live on the external surface of the host (gill parasites are considered external parasites because gills are exposed to the external medium, even in species with enclosed opercula) (Barber *et al.*, 2000). Regarding life cycle, there are parasites with direct life cycle which transmit from one definitive host (the host in which the parasite reaches the adult stage) to another and parasites with complex or indirect life cycle in which the parasite requires at least one intermediate host (the host in which immature stages are present) to complete its life cycle (Barber *et al.*, 2000). The degree of parasite infestation can be expressed as intensity (the number of the parasites from the same species that infest one fish host) in terms of an individual host or as prevalence (The percentage of hostess infested by one or more individual parasite of a certain species) (Bush *et al.*, 1997)

Many parasites adapt to environmental conditions by manipulating fish behavior (make it susceptible to predation) to facilitate the parasite transmission from the fish to the following host of the parasite life cycle (Moore, 2002). In their study on the effect of parasite on fish behavior, Barber *et al.* (2000) reported that parasite infection can change the host fish following behaviors: mate choice, sexual behavior, swimming performance, predator-prey relationships, competitive ability, habitat selection, time budget and foraging ability. The more interesting thing is that parasite infection not only changes the behavior of the fish host but also can change the behavior of the potential fish host by adopting tactics to reduce exposure risk like the avoidance of infected individuals, prey selectivity or even habitat avoidance (Barber *et al.*, 2000). Moreover, parasites have obvious effect on ecosystem energetics and food web, not only as parasite of fish but also as food of fish (i.e. predators eat the fish including its parasites or eat the free-living stages of the parasite)

(Lafferty, 2008). The unexpected indirect effect of parasites is what's argued by Wood and Lafferty (2014) who reported that parasite assemblages is altered by fishing and this have indirect effect that can alter the function of the ecosystem and the services associated with it

Fish parasite are affected by the ecological systems where they are found and vice versa and because it is responsive to variable types of impacts like eutrophication pesticides, heavy metals, wastes (agricultural and industrial) and thermal effluent, it can be used as indicators of environmental impacts and aquatic environmental quality (Lafferty, 2008; Dzika &Wyżlic, 2009). Moreover, trematodes can be used as bioindicators of the abundance of fish communities and diversity, it can also be used to compare biodiversity stat across several locations and to monitor biodiversity changes overtime (Huspeni *et al.*, 2005; Hechinger *et al.*, 2006).

The study of fish health is very important for namely fisheries management and one of the major aspects on fish health is parasites. The rate of parasite infestation determines the damage that is caused by parasites to fish where lightly infected fish will not be affected, heavily infected fish may die or develop damage which may be mechanical (tissue replacement and fusion of gill lamellae), physiological (altered growth, detrimental behavioral responses, immunomodulation and cell proliferation) or reproductive (Iwanowicz, 2011). In spite of the small size of the parasite comparing to the host, its invasion, movement around or growing inside the host affect its biology, which lead to render growth, fitness and survival (Barber *et al.*, 2000).

Some fish parasites infestation can lead to serious economically important effects such as disease outbreaks in infested fish population, nutritional effects that reduce

productivity, serious long-term changes in the structure of the population, spoiling fish tissue that lead to decrease fish market value and reduce the desire for fish as food for consumers (Barber Poulin, 2002).

Some fish parasites can be transmitted to humans and cause diseases especially where the traditional food is lightly salted, cold-smoked, under cooked or raw fish and in some fish production systems where health control approaches are not applied. Indeed, foodborne trematodiases (a group of parasitic infections caused by trematodes) is endemic in some societies (Barber & Poulin, 2002; Khalil *et al.*, 2014; Who, 2017). Allergic reaction can happen when human ingest live or dead larvae of Anisakidae, more than 2000 infections are diagnosed every year in human worldwide, other clinical symptoms of anisakidosis include stomach and oesophagus irritation, vomiting and diarrhea and sometimes severe abdominal and epigastric pain (Pozio, 2013; Buchmann & Mehrdana, 2016).

In spite of the complex factors controlling presence and development of parasite communities in the fish host, they are greatly depending on the use of local habitat, geographic range, diet and size of the fish host; the importance of the fish host body size come from its effect on diet and determining the size of the available space for colonization (Barber & Poulin, 2002).

Khurshid *et al.* (2013) predicted that length, seasonality and gender can affect parasite infestation and their results showed that infection prevalence significantly increase with length of the fish host and positively correlated with season distribution but the gender was negatively correlated with it as male and female fish were equally susceptible to parasite infestation.

Bayoumy et al. (2012) studied parasite-host associations of metazoan parasites with

eight species of fish from Saudi Arabia, for *Acanthgyrus bifasciatus* the infestation prevalence of Monogenea was significantly negatively correlated with weight only.

Ibrahim (2012) studied *Tilapia zillii* parasites infracommunity from Egypt in relation with host origin, sex and length, he found that sex significantly affect intensity and prevalence of infection (females higher than males) and length of the host was positively correlated with intensity and prevalence of infection.

Ramadan (1991) studied the relationship between parasitic infestation and sex, length and season of three species of the fish host *Tilapia* from Egypt and he found, in one hand, that female is heavy infested when compared to males of the same species. In the other hand, large sized fishes are more infested than small ones and winter is the season of higher infestation while summer is the lowest season of infestation, nevertheless he did not make any statistical analysis to test the significance of his results.

Many studies have been done on the parasites of *Acanthopagrus bifasciatus* in the Arabian Gulf and Red Sea, Table 1 shows the parasites encountered during those studies and their localities. While little studies have been done on the parasites of *Pomacanthus maculosus*. Table 2 shows the parasites encountered during those studies and their localities.

 ${\it Table 1 Parasites encountered previously from Acanthopagrus bifasciatus and their locality}$ 

Parasite group	Parasite species	Locality	References
	Lamellodiscus donatellae	Egypt (Red Sea)	Aquaro et al. (2009)
	Polylabris acanthopagri	Egypt (Red Sea)	Aquaro et al. (2009)
	Placodiscus acanthopagri	Egypt (Red Sea)	Aquaro et al. (2009)
Monogenea	-	Saudi Arabia (Arabian Gulf)	Bayoumy et al. (2012)
	Caligus kuwaitensis	Saudi Arabia (Arabian Gulf)	Bayoumy et al. (2013)
	Benedenia acanthopagri	Saudi Arabia (Arabian Gulf)	Hassan <i>et al.</i> (2015)
	Polylabris lingaoensis	Egypt (Red Sea)	Bayoumy <i>et al.</i> (2015)
	Plagioporus sp.	Qatar (Arabian Gulf)	Saoud <i>et al.</i> (1986a)
	Derogenes varicus	United Arab Emirates (Arabian Gulf)	Kardousha (2003)
Trematoda	Macvicaria chrysophrys	Egypt (Red Sea)	Hassanine & Gibson (2005)
	Neowardula brayi	Saudi Arabia (Red Sea)	Al-Jahdali (2010)
	-	Saudi Arabia (Arabian Gulf)	Bayoumy <i>et al</i> . (2012)
Cestoda		Qatar (Arabian Gulf)	Al Kuwari & Kardousha (2002)

<sup>\*</sup>Continued

# \*Continued

Parasite species	Locality	References
Anisakis larvae	United Arab Emirates (Arabian Gulf)	Kardousha (1992)
-	Saudi Arabia (Arabian Gulf)	Bayoumy et al. (2012)
-	-	-
	Anisakis larvae -	Anisakis larvae United Arab Emirates (Arabian Gulf)  - Saudi Arabia (Arabian Gulf)

Table 2 Parasites encountered previously from Pomacanthus maculosus and their locality

Parasite group	Parasite species	Locality	References
Monogenea	-	-	-
Trematoda	-	-	-
Cestoda	Callitetrarhynchus gracilis	Qatar (Arabian Gulf)	Al Kuwari and Kardousha (2002)
Nematoda	Cucullanus extraneus	Iraq (Arabian Gulf)	Li et al. (2016)
Copepoda	-	-	-
Isopoda	-	-	-

The Gulf is a shallow water body located between latitudes 24° and 30° N in the arid subtropics with a maximum length of 990 km and width of 370 km, mean depth of 36 m; it is connected by the Strait of Hormuz to the Gulf of Oman (Torquato *et al.*, 2017).

Fisheries are considered the most important natural resource after Oil in the Arabian Gulf countries (Carpenter *et al.*, 1997). Bayoumy *et al.* (2012) studied metazoan parasites of Saudi Arabia and they found that *Acanthopagrus bifasciatus* was found infested by Monogenea (prevalence 50%) Crustacea (prevalence 50%) and Digenea (prevalence 40%).

In United Arab Emirates, Grandcourt *et al.* (2004) studied the biology and stock assessment of *Acanthopagrus bifasciatus*, while Taher (2010) studied the biological aspects of the same fish including spawning season, sex ratio and food habits and, later on Grandcourt *et al.* (2010) studied the demographic parameters and life history of five fish species including *Pomacanthus maculosus*.

Hassan *et al.* (2015) studied the monogenean infection of cage-cultured three fish species in a private fish farm in Saudi Arabia and he found that *Acanthopagrus bifasciatus* is infested (22.91 %) by *Benedenia acanthopagri* and the infestation rate was higher in summer and lower in winter.

Kardousha (2003) re-described ten species of trematodes including *Derogenes* varicus infesting the fish host *Acanthopagrus bifasciatus* from United Arab Emirates.

Kardousha (1992) studied 40 fish species from United Arab Emirates for nematode larva and he found that *Acanthopagrus bifasciatus* is infested (25 %) by the nematode larva *Anisakis* sp. Hassan *et al.* (2013) studied *Anisakis* larvae in some marine fishes of Saudi Arabia including *Acanthopagrus bifasciatus* and *Pomacanthus maculosus* and they found five species of fish infested by the parasite but *Acanthopagrus bifasciatus* and *Pomacanthus* 

*maculosus* was free of infestation. In Iraq Li *et al.* (2016) made the first report of a nematode (*Cucullanus extraneus* n. sp.) infesting *Pomacanthus maculosus*.

Bayoumy *et al.* (2013) re-described the copepod parasite *Caligus kuwaitensis* from the gills of *Acanthopagrus bifasciatus* from Saudi Arabia by using light and electron microscopy, the prevalence of the parasite was 38.18 %.

The first record of the monogenean parasite *Polylabris lingaoensis* from the gills of the fish host *Acanthopagrus bifasciatus* from Egypt (Red Sea) was recorded by Bayoumy *et al.* (2015) moreover the authors observed the behavior of the fish regarding swimming and respiration and made clinical examination to record pathological effects of the parasite on the fish host.

The peninsula of Qatar is located in the south of the Arabian Gulf (Central part) in a mid-location between Shatt Al-Arab (in the north) and Strait of Hormuz (in the south). The State of Qatar include the peninsula main land and the islands around it between latitude 24° 27' and 26° 10' North and longitudes 50° 45' and 51° 40' East (Saoud *et al.* 1986; Kardousha, 2016).

El Sayed & Abdel-Bary (1993 & 1994) made a biological study on *Mylio bifasciatus* (a synonym to *Acanthopagrus bifasciatus*) from Qatari water and they concluded that gonadal maturation takes place from December to March and spawning occur in April. The fish reaches sexual maturation at the length of 17.46 cm for males and 19 cm for females and male to female ratio is 2:1. The end of the 1<sup>st</sup> year of life is the highest in length growth then it gradually decreases through the fish lifetime.

Torquato *et al.* (2017) made an assessment of taxonomy, functional diversity, diel migration and vertical distribution of some fish assembling around Al Shaheen oil field in

Qatari waters. They recorded 12822 fishes belonging to 83 taxonomic group (the recorded fishes included *Acanthopagrus bifasciatus* and *Pomacanthus maculosus*). The following notes on the two-fish species were recorded: for *Acanthopagrus bifasciatus* the number of individual was 390 with a frequency of 31.8%, the suggested species' traits were as follows: trophic group: invertivore; body shape: oblong; type of swimming: subcarangiform; motility: roving; diet: mobile benthic invertebrates; vertical distribution: benthopelagic and the depth range: 0 - 10 m. For *Pomacanthus maculosus* the number of individual was 1562 with a frequency of 66.9%. Suggested traits are as follows: the trophic group: omnivore; body shape: oval; type of swimming: subcarangiform; motility: roving; diet: colonil sessil invertibrate; vertical distribution: benthopelagic and the depth range: 21 - 30 m.

A general survey on helminth parasites of fish was performed by Saoud *et al.* (1986a). They examined 462 fishes (33 species belonging to 11 families) and they recorded 18 genera of trematodes for the first time from the Arabian Gulf. The fish host *Mylio bifasciatus* which is a synonym to *Acanthopagrus bifasciatus* was found to be infested with the trematode *Plagioporus* sp. with a prevalence of 7.7 %. The list of infested fish species did not include *Pomacanthus maculosus*.

The trematode genera *Hamacreadium* was re-described by Saoud *et al.* (1986b) from *Lutjanus russelli*. and *L. fulviflamma* and *Cainocreadium* from some fish species.

Later Saoud *et al.* (1987) reviewed the trematode genus *Pseudoplagioporus* and redescribed *P. microrchis* from *Lethrinus lentjan* and *L. nebulosus*.

One new species of trematode *Allacanthochasmus lutjani* was recorded by Saoud *et al.* (1988a) from the fish host *Lutjanus fulviflamma*. The same authors recorded two new species of trematodes *Helicometrina qatarensis* and *Stephanostomum nagatyi* (Saoud *et al.*,

1988b).

Trematodes and Cestodes in 61 bony fish species and 14 cartilaginous species were studied by Al-Kawari *et al.* (1996), they recorded 36 trematode genera and 9 cestode genera. They found that *Acanthopagrus bifasciatus* was infested by two genera of trematode (*Derogenes and Plagioporus*) with a prevalence of 8.8 % while *Pomacanthus maculosus* was not mentioned in the list of infected fish.

Saoud *et al.* (2000) studied trematodes and described new trematode species *Stephanostomum qatarense* but the list of infected fish species did not include *Pomacanthus maculosus* or *Acanthopagrus bifasciatus*.

Al Kuwari et al. (2001) recorded the trematodes Erilepturus hamati and Ectenurus trachuri for the first time from the Arabian Gulf and considered Plectorhynchus sordidus as a new host for Ectenurus trachuri and Alectis indica as a new host for Lecithchirium macrorchis.

Kardousha et al. (2002) studied Monogenea and he recorded *Plectorynchus schotaf* and *Carangoides bajad* as new hosts for *Encotyllabe kuwaitensis* and *Encotyllabe spari* respectively.

Al Kuwari and Kardousha (2002) made a parasitological survey on 47 fish species (most of them economically important) belonging to 42 families, they recorded 35 species of helminths (2 species of Monogenea, 27 species of Digenea, 3 species of Cestodes, Anisakids nematodes and 2 species of Acanthocephala). The study recorded 18 parasites as a new record in the Arabian Gulf. Both *Mylio bifasciatus* (which is a synonym to *Acanthopagrus bifasciatus*) and *Pomacanthus maculosus* were found to be infested with the cestode *Callitetrarhynchus gracilis* with a prevalence of 8% and 30% respectively.

Recently, Kardousha (2016) revised the current status of fish parasites in Qatari waters. He summarized the work which have been done since 1986 (first publication on fish parasite in Qatar) till 2002 (last publication on fish parasite in Qatar) as follows: Qatari water have 136 known fish species 51 species have been investigated for parasites and 46 species belonging to 7 groups of parasites have been recorded (1 Microsporidia, 1 Myxosporea, 5 Monogenea, 33 Trematoda, 3 Cestoda, 1 Nematoda, 2 Acanthocephala and no Crustacea). Finally, he concluded that parasitic crustacea have not been encountered from Qatari fish hosts in the previous studies that constitutes a gap which is addressed in this present study.

Acanthopagrus bifasciatus (Forsskål, 1775) belongs to family of Sparidae, commonly known as seabreams; distributed in the Western Indian Ocean, Red Sea and Arabian Gulf; lives around coral reefs; caught by hand lines and traps and the model size ranges from 20 to 35 cm (maximum 55 cm) (Carpenter et al., 1997; FishBase, 2017a). The spawning season for Acanthopagrus bifasciatus in the Arabian Gulf is during January and February and it feeds on snails, followed by crabs (Taher, 2010). Acanthopagrus bifasciatus is a popular food fish (Grandcourt et al., 2004) and it is used for aquaculture industry (Hassan et al., 2015)

<u>Pomacanthus maculosus</u> (Forsskål, 1775) belongs to the Pomacanthidae family commonly known as angelfishes; distributed in the Western Indian Ocean, Red Sea and Arabian Gulf; lives around rocky and coral reefs; feeds mostly on sponges. *P. maculosus* is caught by handlines, trawls and traps and the maximum size is 55 cm (Carpenter *et al.*, 1997; FishBase, 2017b), nevertheless angelfish is not usually targeted and is caught as bycatch. The spawning season of *P. maculosus* is short and single from September to October

(Grandcourt *et al.*, 2010). In spite that *P. maculosus* is usually discarded from trap catches because it has no market value as food (Grandcourt *et al.*, 2010), it is an ornamental beautiful fish that have a very important commercial value when maintained alive (Li *et al.*, 2016).

Accordingly, studying diversity of fish parasites drew little attention but there is a persistent need for studying fish parasitology as parasites lead to commercial loss in fisheries industry and aquaculture and have implications on human health and socioeconomy in both developed and developing countries (Barber *et al.*, 2000). Nevertheless, more parasite species have been described than ever recently and studies highlighted the need for additional research in this topic (Poulin, 2014) emphasizing the risk of not having enough parasitologist expert in taxonomy to cope with the identification and description of newly discovered species as the number of expert taxonomist is very small and limited to individuals in "later stage of their career" (Poulin, 2014).

"The marine environment in Qatar is historically important and constitutes a cultural symbol and a natural source of food, water and wealth for the people of the country" (GSDP. 2009). The uniqueness of the marine environment of Qatar with its unique hydrography, unusual assemblage of fauna, harsh climate and recent degradation through multiple anthropogenic pressures urge for the specialized study of every component in this rich environment including parasite diversity.

According to the above literature review no recent studies have been done on the diversity of parasitic fauna of Qatari fish populations since 2002. Moreover, parasitic crustaceans have not been studied earlier in Qatar.

### **Objectives of the study**

The general aim of the present work is to study the parasitic diversity in two coralreef associated species (*Acanthopagrus bifasciatus* and *Pomacanthus maculosus*) from Qatari coastal waters with special emphasis to ecto-parasites including crustacean parasites.

The detailed objectives of this study are:

- Identification and description of the encountered parasites from the two fish species and compare with previously studies, especially from Qatar and the Arabian Gulf.
- Description of the anatomical observations in the fish host and discussing if it
  indicates the health state of the fish or it has any relation to the parasitic
  infestation.
- Record the prevalence of the parasites in both fish hosts, describe their microhabitat. Describe the, host specificity and define the possibility of mixed infection.
- Study the relationship between fish size and infestation, their spatial variations and gut contents.

## MATERIALS AND METHODS

## 1. Sampling of fish

A total of 135 individuals of two fish species, *Acanthopagrus bifasciatus* and *Pomacanthus maculosus*, were examined for parasite infestation during the period from March 2016 to March 2017. The fish samples and their corresponding numbers are presented in Table (3).

Table 3 Fish samples examined from Qatari water during the present study and their corresponding numbers

Fish	Acanthopagrus bifasciatus	Pomacanthus maculosus	
species			
Image			
Commo	Twobar seabream	Yellowbar angelfish	
n names		Ç .	
Local	Faskara or Bent El- Nwakhaza	Ghanfouz	
names	فسكره أو بنت النواخذة	غنفوز	
Number	89	46	

The two fish were chosen because they are non-migratory and associated to coral reefs (Carpenter *et al.*, 1997; Grandcourt *et al.*, 2004; Iwatsuki & Heemstra, 2011; Samiei Zafarghandi *et al.*, 2013). *Pomacanthus maculosus* is easy to capture directly from coral reefs and *Acanthopagrus bifasciatus* is available in Doha markets as a local fish (it is not imported from outside Qatar).

Identification of fish was done according to Carpenter *et al.* (1997). Figure 1 shows the notes for the identification of the two species.

Fresh fish samples of *Acanthopagrus bifasciatus* were purchased from Doha whole sale market and from the seafood department of different hypermarkets in Doha.

Fish samples of *Pomacanthus maculosus* were caught by spearfishing from coral reefs only one time the fish was found in the whole sale marked and purchased from there. Figure 2 shows the locations and number of examined fish from each location.

After capture or purchase the fish were immediately placed in ice and brought to the laboratory.

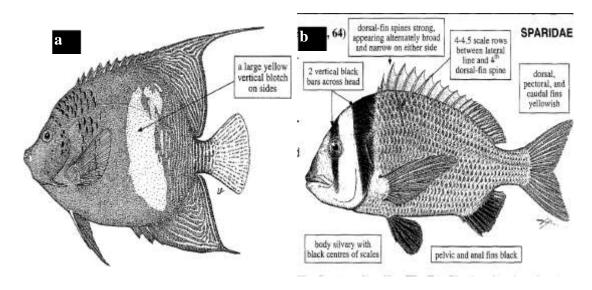


Figure 1. Notes used for the identification of the two species during the present study (Carpenter et al., 1997): a) Acanthopagrus bifasciatus. b) Pomacanthus maculosus.



Figure 2. Sampling locations and number of *Pomacanthus maculosus* sampled in each location from Qatari water.

### 2. Laboratory procedures:

The location and date of collection, length, weight, sex (Okoye *et al.* 2014) and physical health abnormalities of each fish was recorded. Fish was examined and health endpoints were assessed by recording abnormalities in collected fish as: (1) intensive parasitic infections (fish with two species of parasites or with four parasites or more per one fish) (2) ulcers, (3) tumors, (4) fin erosion and (5) body abnormalities (Moore, *et al.*, 1997) Gills, Buccal cavity, eyes and skin were carefully investigated to reveal ectoparasites such as Monogenea, and Crustacea.

The digestive system of each fish was dissected and the gut contents was separately washed-off in 0.75% saline solution, and then checked using a stereo-microscope for the presence of parasites.

Gut contents (food) for each fish was examined, recorded and kept (if needed) in 70% ethyl alcohol for further investigation.

Body cavity, eyes, liver, gall bladder, gonads and kidney also were separately investigated for the presence of any parasite.

Encountered parasites were transferred to clean watch glasses containing 0.75% saline solution using fine forceps or brush. The contents of each watch-glass were transferred to small glass bottles containing saline solution, and then vigorously stirred to get rid of debris. Parasites were transferred again to Petri dishes containing clean 0.75% physiological saline solution.

The parasite samples were divided into two groups. One group was fixed for drawing and identification; the second group was fixed in 2.5% glutaraldehyde for possible scanning electron microscopy studies.

## **Techniques employed in the present study:**

### 1. Whole mount preparations (Hoffman, 1999 and lasee, 2004).

#### 1.1. Monogenea and Trematoda

#### 1.1.1. Fixation:

Worms were washed and left few minutes in cold saline solution until they become completely relaxed. Specimens were compressed by delicate pressure between two slides.

The slides which contain the flattened parasites were fixed in large Petri-dish containing hot formal acetic acid (F.A.A.) solution (10 ml formalin + 5 ml glacial acetic acid + 25 ml ethyl alcohol + 50 ml distilled water + 10 ml glycerol) for two hours. Then the specimens were removed from the fixative and kept in 70 % ethyl alcohol until the time of staining.

### 1.1.2. Staining:

Prior to staining, the parasites were washed several times in water to remove the excess of fixative. The worms were stained using acetic acid-alum carmine staining solution, which was prepared from:

Carmine 25 gm.

Glacial acetic acid 25 ml.

Potassium alum 25 gm.

Distilled water 1000 ml.

The staining process was checked at short intervals under a stereo-microscope until the desired degree of staining is attained.

The stained parasites were rinsed in acid alcohol (freshly prepared by adding a drop of concentrated Hydrochloric acid into 10 ml of 70% alcohol) and checked at short intervals under a stereo-microscope until the perfect staining level reached and the stained parasites

become well differentiated.

## 1.1.3. Dehydration:

Specimens were dehydrated in ascending grades of ethanol (70%, 80%, 90%, 95% and 100%), 10 minutes each.

### 1.1.4. Clearing:

Specimens were cleared in xylene.

## 1.1.5. Mounting:

Finally, specimens were mounted on clean glass slides and permanently covered with thin glass cover using DPX as a mountant.

### 1.2. Nematodes and Crustacea

#### 1.2.1. Fixation:

Nematodes were fixed in warm (60 °C) 70% ethanol. Then they were kept in 70 % alcohol containing 5 % glycerol. Crustacea were fixed in 70 % alcohol.

## 1.2.2. Mounting:

Parasites were mounted on a slide with few drops of lactophenol. An objective lens was used to examine the anterior and caudal ends.

Lactophenol consists of:

Phenol 10 ml
Lactic acid 10 ml
Glycerol 10 ml
Distilled water 10 ml

# 1.3. Drawing and identification:

Hand drawings were made from previously prepared whole mount specimens using the microscope projector. Identification of parasites was done by comparing the specimens with previously described related species (All references are written in detail in the discussion of each parasite).

### 1.4. Images and measurements:

Images and measurements were taken by using AxioCamERC camera, Leica application suite (LAS) software V4 and imagej software.

# 2. Preparation of specimens for scanning electron microscopy (Arafa, 2011):

### 2.1. Fixation:

Specimens were fixed at 4 °C in 2.5% glutaraldehyde

### 2.2. Washing:

Washing was carried out several times in 70% alcohol.

### 2.3. Dehydration:

After complete removal of excess glutaraldehyde, the specimens were dehydrated in ascending grades of ethanol series. Samples were then mounted on the holder and left to dry in the oven, overnight, at 37 °C.

# 2.4. Coating:

Samples were sputter coated with gold and viewed with scanning electron microscope in the electron microscopy unit (Qatar University).

# 3. Statistical analysis:

Statistical analysis was done using IBM SPSS statistics Version 23

CHAPTER 2: PARASITES ENCOUNTERED DURING THE PRESENT STUDY

There were eight species of parasites encountered during the present study: five

ecto-parasites (Polylabris mamaevi, Alella sp., Caligus haemulonis, Lernanthropus sarbae

and Pranzia larva of Gnathia africana) and three endo-parasites (Stephanostomum sp.,

Dujardinascaris sp. and Cucullanus sp.).

Description of the encountered crustacean parasites in comparison with previously

described related species was mentioned in detail in this chapter because the present study

is the first record of crustacean parasites in Qatar.

Parasites other than crustacea (Monogenea, Trematoda and Nematoda) were

identified but the detailed of their description and comparison is not mentioned in the

chapter.

Caligus haemulonis Krøyer, 1863

Classification.

**Kingdom:** Animalia

Phylum: Arthropoda

Subphylum: Crustacea Brünnich, 1772

Class: Maxillopoda Dahl, 1956

Subclass: Copepoda Milne-Edward, 1840

**Order:** Siphonostomatoida Thorell, 1859

Family: Caligidae Burmeister, 1834

Genus: Caligus O. F. Müller, 1785

23

### **Prevalence:**

Five out of the 89 fish (*Acanthopagrus bifasciatus*) examined (5.6%) were found to be infested by *Caligus haemulonis*. The parasite was found only on the gills, ranging in number from one to three individuals. The parasite was found alone or in a mixed infestation with *Polylabris mamaevi*. The parasite was usually associated with excessive mucous secretion and strange branches in the gills (Table 4).

Table 4 Prevalence of Caligus haemulonis found in the host species Acanthopagrus bifasciatus sampled from Qatar.

					Number	
	No. of	No. of	Prevalenc	Microhabita	of	Mixed
Host species	examine	infeste	e	t	parasite	infestatio
	d fish	d fish	C	ι	s per	n
					fish	
Acanthopagru						Polylabri
	89	5	5.6%	Gills	1 - 3	S
s bifasciatus						mamaevi

# **Description of adult female Based on 5 females (measurements in table 4):**

The body (like all in the Caligid family) consists of four parts (tagma): Cephalothorax, fourth leg bearing segment, genital complex and abdomen (Fig. 3 &4).

### 1) Cephalothorax:

The cephalothorax (Fig. 5 & 6) has a unique structure in Caligid copepods as they use it as a sucker-like attachment organ. The ventral cephalothorax contains attachments and feeding appendages, while the dorsal is roughly rounded and smooth. The cephalothorax shield is wider than long (sub-orbicular in shape).

At the ventral surface of the frontal plate there is a pair of prominent lunules (cuplike suckers, Fig. 5), which are finely striated and the distance between them is less than their diameter. The siphonostomatoid buccal apparatus (siphon or mouth cone, Fig. 5 and Fig. 10) has a typical tubular structure and is carried in a folded position, parallel to body axis. On the median surface between the two lunules there is a half circular structure (rugose or sucker apparatus, Fig. 5) with a central sulcus.

The first antenna (antennule, Fig. 3 & 4) is small, without distinguishing characteristics and located on the lateral sides of the frontal plate. The second antenna (antenna Fig. 3 & 5) has a curved (right-angle) pointed spine (claw). The postantennal process (Fig. 3 & 5) has a sharply pointed (right-angle) claw, bearing basal papilla carrying 3 setules.

The First maxilla (maxillul, Fig. 3) has a small con-shaped process with papilla on the anterior internal corner. The second maxilla (maxilla, Fig. 3 and Fig. 9) is composed of two segments: the proximal one is cylinder and unarmed, while the distal one is armed with two pinnate setae. The third maxilla (maxilliped, Fig. 3 & 5) consists of two segments: the proximal one is large, wide and conical shaped, while the distal one is armed with a large pointed claw. The sternal furca (Fig. 3 & 5) has two curved tines, which are slightly longer than the subquadrat box.

The first leg (Fig. 3, 6 and 10) has a cylindrical protopod (coxopod) that has papilla with 3-setules on the lateral side. Above the papillae there is a patch of tiny spinules and, on the terminal part of the protopod, there is a triangular process, the exopod, consists of two segments. The first segment is wide, the posterior edge fringed with a row of small setules and the anterior edge having pointed process with lateral setule. The second segment is smaller and armed with four pinnate plumose setae. Medial margin without the usual three setae.

The second leg (Fig. 3, 7 and 9) has a small coxa with a large lateral plumose seta. The exopod consists of three segments: the first and second ones have lateral plumose seta at the inner side and a lateral large spine which bent internally at the outer side, while the third segment has a small spine and 7 plumose setae (four long, one medium and one small seta bent to the outside on the lateral outer edge of the segment). The endopod consists of three segments: the first and second segments have one lateral plumose seta at the inner side and a small spine at the outer side, while the third segment has six long plumose setae.

## 2) Fourth leg bearing segment:

It is wider than long, free and located between the cephalothorax and the genital complex. It does not attach to any other limbs or appendages. The exopod of fourth leg (Fig. 3 and 7) is two segmented: the first segment has a long spine on the outer distal corner, while the second segment has four spines, three large on the terminal part and one medium on the outer margin.

### 3) Genital complex:

It is globose (sometimes heart-like, Fig. 3, 4 and 8), longer than wide. Sometimes there are a pair of semi-oval spermatophores attached to ventro-posterior surface of genital

complex. The egg-sac is less one-half of body length (Fig. 11), containing 18 – 24 (21) egg.

4) Abdomen:

It is small, longer than wide, two-segmented, attached anteriorly to the genital complex and ending with two terminal uropods (caudal ramus). The caudal ramus (Fig. 3, 4 and 8) are distinct from the abdomen, flat and slightly longer than wide, bearing one short and three long setae. The short is not pinnate and locate in the posterior-lateral corner while the long are pinnate and plumose on the posterior margin.

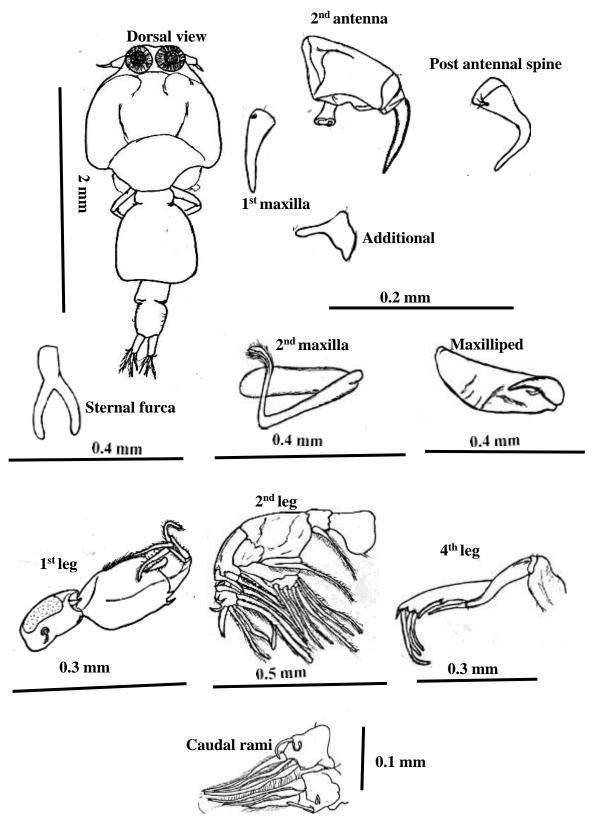


Figure 3. Adult female Caligus haemulonis and its enlarged appendages.

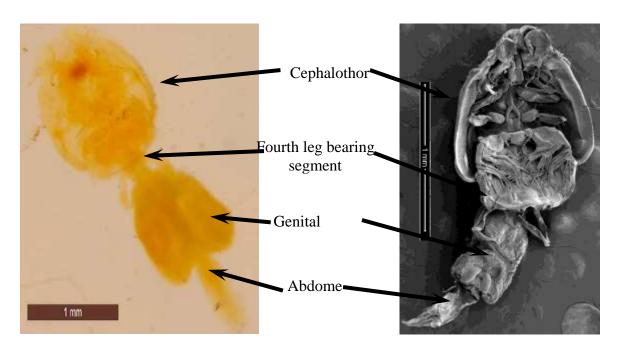


Figure 4. Ventral view of the adult female Caligus haemulonis showing the four body parts a) light microscopic photomicrograph. b) SEM.

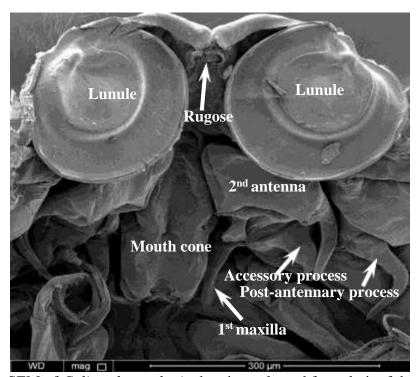


Figure 5. SEM of Caligus haemulonis showing enlarged frontal pit of the cephalothorax.

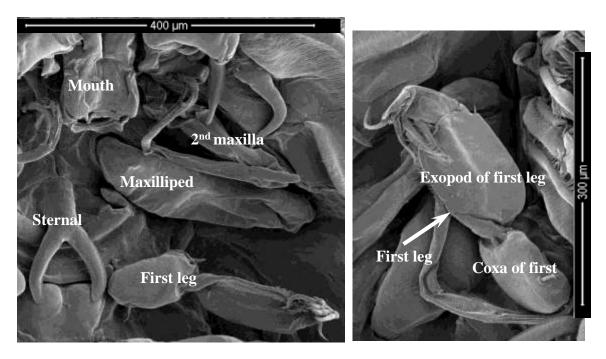


Figure 6. SEM of Caligus haemulonis showing enlarged cephalothorax

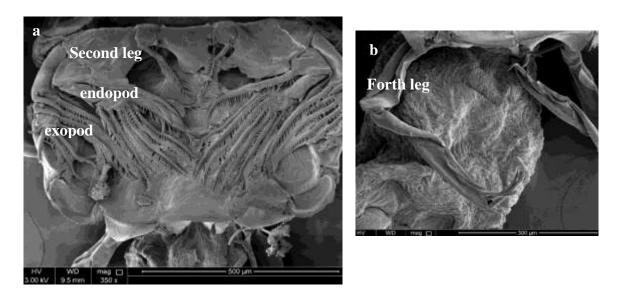


Figure 7. SEM of Caligus haemulonis a) Second leg. b) Fourth leg.

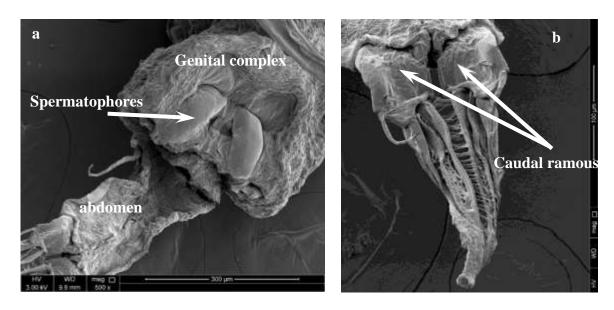


Figure 8. SEM of Caligus haemulis a) Enlarged genital complex. b) Enlarged uropod.

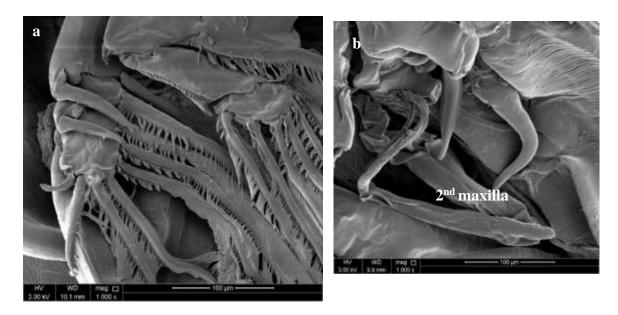


Figure 9. SEM of Caligus haemulonis a) Enlarged 2<sup>nd</sup> leg. b) Enlarged 2<sup>nd</sup> maxilla

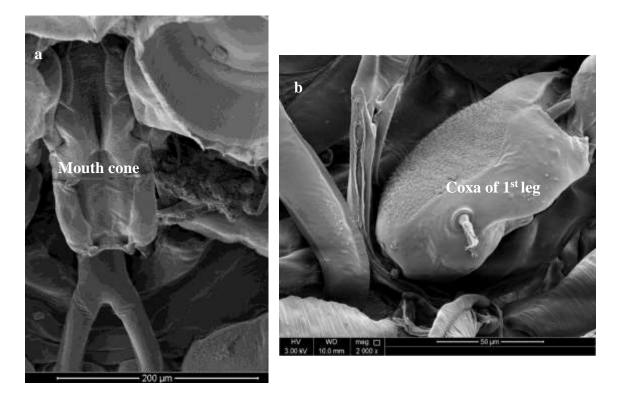


Figure 10 SEM of Caligus haemulonis a) Enlarged mouth cone. b) Enlarged coxa of  $1^{st}$  leg

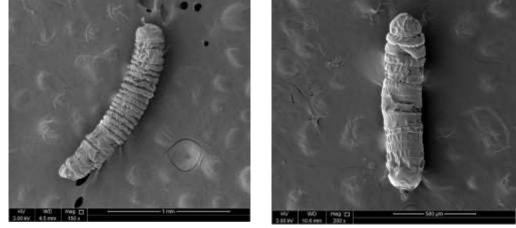


Figure 11. SEM of Caligus haemulonis egg sac.

#### **Discussion:**

Caligids (usually referred to as sea lice) are widespread parasite in marine and brackish waters as they have good tolerance to temperature, salinity, biochemical compounds and can infest wide range of hosts and switch between hosts due to the presence of host-finding behaviour (Boxaspen, 2006, Mordue & Birkett, 2009 and Muhd-Faizul, 2013). Caligids can harm their hosts by their mechanism of attachment or by feeding on their host blood, mucous and tissue which cause retardation in fecundity, growth or even survival depending on the severity of infection, fish size, fish age and the general health state of the fish (Johnson *et al.*, 2004)

As described by Muhd-Faizul (2013) the life cycle of Caligids starts with the implementation of the spermatophores of the male on the female genital complex, which develop into an egg sac. The eggs hatch into the planktotrophic naupli that evolve into the infective larval stage (copepodid), which attach to the host and transform into chalimus then to pre-adult and finally evolves into adult.

The genus *Caligus* was established by Müller in 1785 and has grown rapidly to contain more than 250 species (Ho *et al.* 2000).

Kroyer (1863) described *Caligus haemulonis* from *Haemulon elegans* from the Danish West Indies. Cressey (1991) and Boxshall & El-Rashidy (2009) described the same species from marine fish in Florida and Brazil respectively. Recently, Suárez-Morales *et al.* (2010) redescribed *Caligus haemulonis* from the gills of *Haemulon sciurus* in Mexico.

In the Arabian Gulf, Kabta and Tareen (1984) first described *Caligus kuwaitensis* as a new species from the skin of *Plotossus anguillaris*, in Kuwait Bay. Bayoumy *et al.* (2013) reported *Caligus kuwaitensis* from gills, buccal cavity, pharyngeal cavity and mouth

of Acanthopagrus bifasciatus in Dammam, Saudi Arabia.

Recently, Mahdy and Abu El Ezz (2015) described female *Caligus kuwaitensis* from the gills of marine fish of Egypt.

During the present study, all the encountered parasites were without egg sac and this was confusing because in some cases egg sacs was found alone without the parasite itself. This may be explained by the findings of Schram (2000), who stated that there is a hook apparatus on the genital complex of caligids that is under muscular control of the parasite and is responsible for the release of the egg sac after hatching. Nevertheless, if stressed, the female can release all the egg sacs with the fertilized eggs before hatching.

All the encountered parasites were found to be female, as the antenna (second antenna) have a sharp point claw and this is a distinguished character between male and female (Cressey, 1991; Boxshall & El-Rashidy, 2009 and Suárez-Morales *et al.*, 2010). Table (4) shows a comparison between female *Caligus haemulonis* of the present study and previously described related species.

By comparing the present parasite with previously described related species the following can be concluded:

1) The measurements of the body length, cephalothorcic shield, genital complex of the present parasite is in the range of *Caligus haemulonis* described by Cressey (1991), Boxshall & El-Rashidy (2009) and Suárez-Morales *et al.* (2010); while they are obviously larger than *Caligus kuwaitensis* described by Kabta & Tareen (1984), Bayoumy *et al.* (2013) and Mahdy & Abu El Ezz (2015).

- 2) The lunules of the present parasite are larger than those of *Caligus kuwaitensis* (Bayoumy *et al.*, 2013).
- 3) The shape of the rugose is different from *Caligus kuwaitensis* (Mahdy & Abu El Ezz, 2015).
- 4) The abdomen of the present parasite is 2-segmented and this is in agreement with *Caligus haemulonis* described by Cressey (1991), Boxshall & El-Rashidy (2009) and Suárez-Morales *et al.* (2010). On the contrary, the abdomen of *Caligus kuwaitensis* is unsegmented, as described by Kabta & Tareen (1984), and Mahdy & Abu El Ezz (2015). Only Bayoumy *et al.* (2013) describe the abdomen of *Caligus kuwaitensis* as 2- segmented and this is disagreeing with the description of Kabta & Tareen (1984) who was the first one to describe the species.
- 5) The caudal ramus of the present parasite is much larger than *Caligus kuwaitensis* described by Kabta & Tareen (1984), Bayoumy *et al.* (2013) and Mahdy & Abu El Ezz (2015) that described it as much reduced or very small.
- 6) The structure of the antenna (second antenna) is similar to *Caligus haemulonis* Cressey (1991) and Boxshall & El-Rashidy (2009).
- 7) The presence of additional process in the present parasite resemble *Caligus haemulonis* (Boxshall & El-Rashidy, 2009).
- 8) The structure of the first leg of the present parasite resembles *Caligus haemulonis* described by Cressey (1991), Boxshall & El-Rashidy (2009) and Suárez-Morales *et al.* (2010), in having the medial margin of the exopod distal segment without the usual three setae which is considered a distinguishing

character for the Products-group of Caligids (Boxshall & El-Rashidy, 2009) to which belongs *Caligus haemulonis*. On the contrary, *Caligus kuwaitensis* described by Kabta & Tareen (1984), Bayoumy *et al.* (2013) and Mahdy & Abu El Ezz (2015), the medial margin of the exopod distal segment have the usual three setae.

9) The structure of leg four of the present parasite resembles to *Caligus haemulonis* described by Cressey (1991), Boxshall & El-Rashidy (2009) and Suárez-Morales *et al.* (2010), in having the first segment of the exopod with a long spine on the outer distal corner while the second segment has four spines, three large on the terminal part and one medium on the outer margin.

For all the previous remarks the present parasite can be considered as *Caligus haemulonis* (Krøyer, 1863) and according to the available data this is the first record of the parasite, not only in Qatari waters but also in the Arabian Gulf. Moreover, *Acanthopagrus bifasciatus* can be considered as a new host for the parasite.

The appearance of *Caligus haemulonis* in the Arabian Gulf for the first time can be explained by the notice of Muhd-Faizul (2013). This author stated that the temperature dependent life cycle of Caligus (i.e., increasing water temperature will shorten the life cycle and increase the abundance), can potentially affect the geographical distribution of the genus and bring new species into new areas (Muhd-Faizul, 2013).

Table 5 Comparison between female Caligus haemulonis of the present study and previously described related species (all measurements

# \*Continued

	Co	aligus kuwaiter	isis		Caligus ha	emulonis	
Character	Kabta & Tareen (1984)	Bayoumy et al. (2013)	Mahdy & Abu El Ezz (2015)	Cressey (1991)	Boxshall & El- Rashidy (2009)	Suárez-Morales <i>et al.</i> (2010)	Present work
Body length excluding setae on caudal rami	2.36 – 2.60 (2.51)	2.00 – 2.42 (2.2)	2.25 - 3.12 (2.5)	3.56	2.96–3.92	3.14 - 3.27	3.2 - 3.49 (3.3)
Cephalothora cic shield	1.36 – 1.40 (1.39) X 1.40 – 148 (1.43)	0.75 – 1.14 (0.93) X 0.84 – 1.22 (0.97)	1.28-1.39 (1.31) X 1.22- 1.35 (1.20)	-	-	1.31–1.37 X 1.40–1.43	1.59 - 1.65 (1.6) X 1.19 - 1.32 (1.3)
Lunules	-	0.279	Finely striated and the distance between them larger than their diameters	-	-	-	0.23 – 0.24 (0.235)
Rugose	-	-		-	-	-	

<sup>\*</sup>Continued

	Co	aligus kuwaiter	isis		Caligus ha	emulonis	
Character	Kabta & Tareen (1984)	Bayoumy et al. (2013)	Mahdy & Abu El Ezz (2015)	Cressey (1991)	Boxshall & El- Rashidy (2009)	Suárez-Morales <i>et</i> al. (2010)	Present work
Mouth cone	-	-		-	-	-	
Genital complex	0.80 – 0.96 (0.87) X 0.72	Trapezoidal 0.67–0.75 (0.71) X 0.58– 0.65 (0.62)	trapezoidal, 0.99-1.07 (1. 01) X 0.68- 0.79 (0.7)	-	-	-	Globose, 0.97 - 0.99 (0.98) X 0.71 - 0.88 (0.80)
Abdomen	Unsegmented 0.08 – 0.12 (0.11) X 0.12 – 0.24 (0.17)	Indistinctly 2- segmented	Unsegmented 0.61-0.73 (0.68) X 0.30– 0.48 (0.3 2)	-	2-segmented	-	2- segmented 0.58 - 0.79 (0.69) X 0.27 - 0.34 (0.30)
Caudal ramus	Much reduced	0.085 X 0.070	0.088 X 0.073	small	-	-	0.24

<sup>\*</sup>Continued

	Co	aligus kuwaiter	isis		Caligus ha	emulonis	
Character	Kabta & Tareen (1984)	Bayoumy et al. (2013)	Mahdy & Abu El Ezz (2015)	Cressey (1991)	Boxshall & El- Rashidy (2009)	Suárez-Morales <i>et</i> al. (2010)	Present work
Female	0	-		-			
Spermatopho re	-	-	OD	-	-		
Second antenna	2	an2 mc pap	-			(C)	
Additional process	-	-	-	-		-	

<sup>\*</sup>Continued

	Co	aligus kuwaiter	ısis		Caligus ha	emulonis	
Character	Kabta & Tareen (1984)	Bayoumy et al. (2013)	Mahdy & Abu El Ezz (2015)	Cressey (1991)	Boxshall & El- Rashidy (2009)	Suárez-Morales <i>et</i> al. (2010)	Present work
Post antennal spine	-	-	45			C	
1 <sup>st</sup> maxilla	2	-			V	-	
2 <sup>nd</sup> maxilla	-	-		-	-	-	
Maxilliped	Ċ	-	B	-		25°	
Sternal furca	8 B	-	9				

<sup>\*</sup>Continued

	Co	aligus kuwaiter	ısis		Caligus ha	emulonis	
Character	Kabta & Tareen (1984)	Bayoumy et al. (2013)	Mahdy & Abu El Ezz (2015)	Cressey (1991)	Boxshall & El- Rashidy (2009)	Suárez-Morales <i>et</i> al. (2010)	Present work
Leg 1	Post	-	tegt excel			E C	
Leg 2		-		or Aller	-	=	
Leg 3		-	-		-		-

<sup>\*</sup>Continued

	Co	aligus kuwaiter	isis		Caligus had	emulonis	
Character	Kabta & Tareen (1984)	Bayoumy et al. (2013)	Mahdy & Abu El Ezz (2015)	Cressey (1991)	Boxshall & El- Rashidy (2009)	Suárez-Morales <i>et</i> al. (2010)	Present work
Leg 4	12	-					
Caudal rami	14 14 18 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1	-	-		-		
Egg-sac	-	less one- half of body length, containing 4 eggs	-	-	-	-	0.9 – 1.57 (1.2) 18 – 24 (21) egg
Host	Plotossus anguillaris	Acanthopag rus bifaciates	Pagrus pagrus	-	Micropogon furnieri	Haemulon sciurus	Acanthopag rus bifaciates
Prevalence	-	38.18%	-	-	-	13 %	5.6%
Intensity	-	-	-	-	-	1 - 2	1 - 3

<sup>\*</sup>Continued

# \*Continued

	C	aligus kuwaiter	isis		Caligus ha	emulonis	
Character	Kabta & Tareen (1984)	Bayoumy <i>et al.</i> (2013)	Mahdy & Abu El Ezz (2015)	Cressey (1991)	Boxshall & El- Rashidy (2009)	Suárez-Morales <i>et al.</i> (2010)	Present work
Microhabitat	Skin	Gills, buccal cavity, pharyngeal cavity and mouth	-	-	-	Gills	Gills
Locality	Arabian Gulf, Kuwait	Arabian Gulf, Saudi- Arabia	Egypt	Florida	Brazil	Mexico	Arabian Gulf, Qatar

# Alella sp. Adday, 2013.

### **Classification:**

**Kingdom:** Animalia

Phylum: Arthropoda

Subphylum: Crustacea Brünnich, 1772

Class: Maxillopoda Dahl, 1956

**Subclass:** Copepoda Milne-Edward, 1840

Order: Siphonostomatoida Thorell, 1859

Family: Lernaeopodidae Milne Edwards, 1840

Genus: Alella Leigh-Sharpe, 1925

## **Prevalence:**

Four out of the 89 fish (*Acanthopagrus bifasciatus*) examined (4.5%) were found to be infested by *Alella* sp. The parasite was found only on the gills, range in number from one to two individuals. The parasite was found alone or in a mixed infestation with *Polylabris mamaevi* and usually combined with excessive mucous secretion on the gills (Table 6).

Table 6 Prevalence of *Alella* Sp. found in the host species *Acanthopagrus bifasciatus* sampled from Qatar.

Host species	No. of examine d fish	No. of infeste d fish	Prevalenc e	Microhabita t	Number of parasite s per fish	Mixed infestatio
Acanthopagru s bifasciatus	89	4	4.5%	Gills	1 - 2	Polylabri s mamaevi

### Description of adult female Based on 5 females (measurements in Table 6):

The body is white creamy in colour divided into three parts (Fig. 12, 13 & 14): Cephalothorax, reduced maxilla and trunk.

Cephalothorax (Fig. 13 & 16) is elongate with lobular laterally projection (aliform process) at the junction between the cephalothorax and the trunk. The posterior aliform process is narrow from one side and have a rounded swelling from the other side.

Maxilla (Fig. 13) is short, inserted between the base of the cephalothorax and the aliform process (Fig. 12 and 13). It has two (collar-shape) lobes nearly equal in size and smaller than the aliform process acting as a base to the insertion of the bulla. Bulla is stout, slender and long with longitudinally striated chitin.

Trunk is pyriform (Fig. 13, 14 & 17), elongate and having a distal rounded small genital process. Egg sac is wide and oval with rounded end (Fig. 13 & 17).

### **Discussion:**

Lernaeopodidae (Osslon, 1869) is a family of siphonostomatoid copepods that has a unique adapted maxilla which work as an anchor-like attachment organ to connect the bulla with the body of the parasite and it contains 49 genera (Adday, 2013).

The harmful effect of *Alella* on the fish (Roubal, 989) is greatly affected by the size of the parasite and the histopathological caused by the penetration of the parasite to the fish host include, oedema and hyperplasia. Chronic inflammation and fusion of the filament is caused by the adult parasite. Bulla penetration to the connective tissue of the fish stimulate chondrocyte proliferation and fibroplasia to enclose the bulla which is separated from the surrounded connective tissue by a tri-laminar wall.

As described by Kawatow et al. (1980) the female life cycle of Allela consists

of seven stages: one nauplius, one copepodid, four chalimus, and the adult stage, but the male life cycle is not well known and the dwarf male carried on the female cephalothorax and this may explain the presence of female only in the present study. The life cycle described by Huyes (2014) is little differ in the presence of the pupa stage after the copepodid stage. The sexual dimorphism takes place in the pupa stage and it is followed by four chalimus stages, young then adult in case of female, while it is followed by one chalimus stage, preadult then dwarf adult in case of male.

The genus *Alella* Leigh-Sharpe, 1925 was established to separate the Clavellodes (Wilson, 1915) that contain aliform extensions at the base of the cephalothorax from other members of the genus (Hewitt & Blackwell, 1987). It consists of seven species (Dippenaar, 2016) and characterized by the presence of aliform process and the reduced maxilla which is consist of two lobes at the base of the cephalothorax and act as a base to the bulla insertion (Adday, 2013).

Alella tarakihi Hewitt & Blackwell, 1987 was described from the gills of the tarakihi Cheilodactylus macropterus from two locations around New Zealand. Alella gibbos Van Niekerk and Oliver, 1995 was described from the tips of gill filaments of Rhabdosargus sarba from Lake St. Lucia, South Africa.

Alella macrotrachelus Byrnes, 1988 was described from the tip of the gill filaments of the Australian Bream Acanthopagrus butcheri, A. australis, A. berda and A. latus from Australia.

In the Arabian Gulf *Alella* sp. (Adday, 2013) was described from the gills of *Acanthopagrus arabicus* from Iraq.

By compairing (Table 7) female *Alella* sp. of the present study and *Alella* sp. (Adday, 2013). the following can be concluded:

- The measurements of cephalothorcic, trunk and maxilla are little larger in the present parasite while the measurements of genital process and egg sac are little smaller.
- 2) The measurements of the bulla is recorded in the present study while it is not mentioned in Adday (2013).
- 3) The fish host in the present study was *Acanthopagrus bifasciatus* while it was *A. arabicus* in Adday (2013).
- 4) The prevalence of the of the parasite in the present study was 4.5% and this nearly like that of Adday (2013) which was 3.6%.
- 5) The intensity of parasite infection in the present study was 1 -2 female while the mean was 0.8 female in Adday (2013).
- The maxilla of the present parasite has a bulla that help to attach the parasite to the fish host so it belongs to the family Lernaeopodidae (Osslon, 1869) and it contain aliform process that characterize the genus *Alella* (Leigh-Sharpe, 1925) but the details of the cephalothorax (which distinguish between different *Alella* species) is not obvious enough to know the species of the parasite.

The mode of attachment is confirmed in the present study by finding some of the parasites firmly attached to the host by sucking the gill filament of the fish inside the bulla of the parasite (Fig. 15).

According to the available data this is the first record of *Alella* sp. in Qatar. The parasite is detected before from *Acanthopagrus butcheri*, *A. australis*, *A. berda A. latus*, and *A. arabicus* but not from *Acanthopagrus bifaciates* which consider a new host for the parasite in the present study.

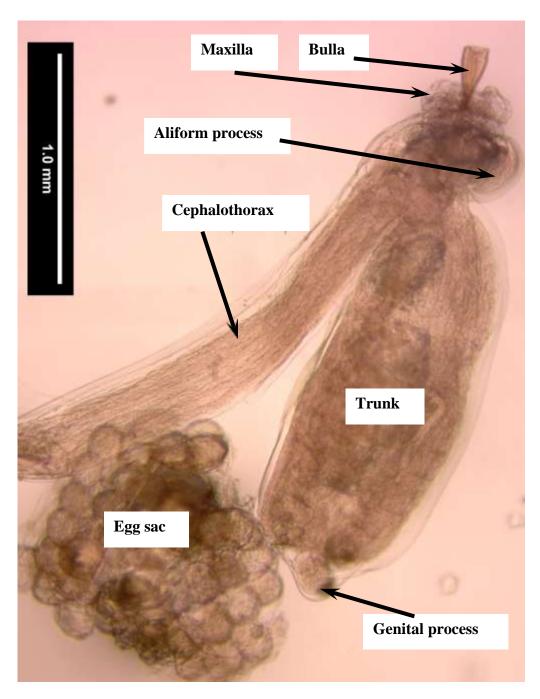


Figure 12. Light microscopic photomicrograph showing lateral view of the adult female Alella sp. with the egg sac bent forward

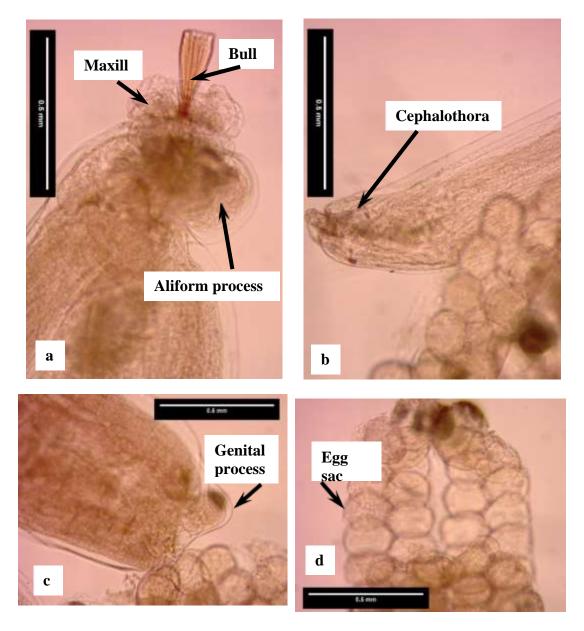


Figure 13. Light microscopic photomicrograph of the adult female Alella sp. showing enlarged parts of: a) anterior part b) anterior tip of cephalothorax c) posterior part d) egg sac

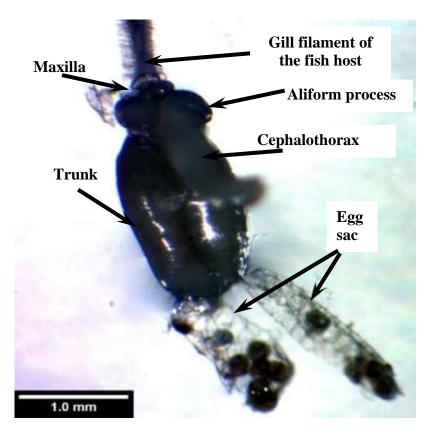


Figure 14. Light microscopic photomicrograph showing ventral view of the adult female Alella sp.

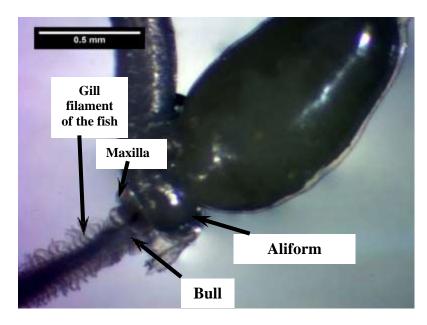


Figure 15. Light microscopic photomicrograph of the adult female Alella sp. showing the mode of attachment of the parasite to the fish host.

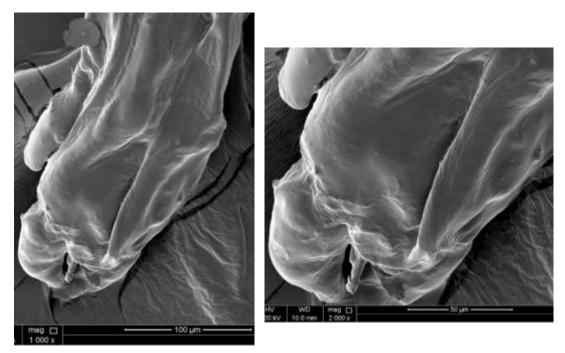


Figure 16 SEM of the adult female Alella sp. showing enlarged terminal part of cephalothorax.

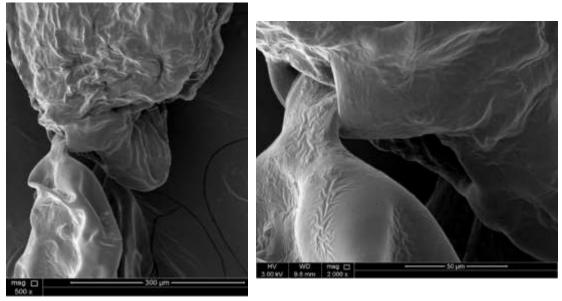


Figure 17. SEM of the adult female Alella sp. showing enlarged terminal part of the body with the egg sac.

Table 7 Comparison between female Alella sp. of the present study and Alella sp. (Adday, 2013). (all measurements are in mm):

Character	Alella sp. (Adday, 2013)	Alella sp. (The present work)
Figure		
Cephalothorax	1.92 - 2.08 (2.0) X 0.26 - 0.32 (0.29)	2.13 – 2.50 (2.32) X 0.36 – 0.42 (0.3
Trunk	Pyriform 1.20 - 1.33 (1.25) X 0.74 - 0.82 (0.78)	1.48 – 1.80 (1.64) X 0.70 – 0.75 (0.7
Maxilla	0.14 - 0.18 (0.15) X 0.28 - 0.32 (0.29)	0.17 – 0.20 (0.19) X 0.27 – 0.30 (29
Bulla	-	0.20 – 0.32 (26) X 0.10 – 0.13 (11.5
Genital process	0.16 - 2.0 (0.17) X 0.12 - 0.16 (0.14)	0.12 - 0.16 (0.14) X 0.19 - 0.20 (0.20

<sup>\*</sup>Continued

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Character	Alella sp. (Adday, 2013)	Alella sp. (The present work)	
Egg sac	2.13 - 2.48 (2.27) X 0.29-0.40 (0.33)	1.68 – 1.98 (1.83) X 0.39 – 0.49 (0.44)	
Host	Acanthopagrus arabicus	Acanthopagrus bifaciates	
Prevalence	3.6 %	4.5 %	
Intensity	Mean 0.8	1 - 2	
Microhabitat	Gills	Gills	
Locality	Arabian Gulf, Iraq	Arabian Gulf, Qatar	

# Lernanthropus sarbae Kensley and Grindley, 1973

### **Classification:**

Kingdom: Animalia

**Phylum:** Arthropoda

Subphylum: Crustacea Brünnich, 1772

Class: Maxillopoda Dahl, 1956

**Subclass:** Copepoda Milne-Edward, 1840

Order: Siphonostomatoida Thorell, 1859

Family: Lernanthropidae Kabata, 1979

Genus: Lernanthropus Blainville, 1822

### **Prevalence:**

Two out of the 89 fish (*Acanthopagrus bifasciatus*) examined (2.2 %) were found to be infested by *Lernanthropus incilis*. The parasite was found only on the gills and only one individual was found on each fish. The parasite was found in a mixed infestation with *Polylabris mamaevi* and usually combined with excessive mucous secretion on the gills (Table 8)

Table 8 Prevalence of *Lernanthropus sarbae* in the host species *Acanthopagrus bifasciatus* sampled from Qatar.

Host species	No. of examined fish	No. of infected fish	Prevalence	Microhabitat	Number of parasites per fish	Mixed infestation
Acanthopagrus bifasciatus	89	2	2.2%	Gills	1	Polylabris mamaevi

### **Description of adult female Based on 2 females (measurements in table 8):**

The parasite is white creamy in colour and the body consists of: head, neck, trunk and urosome (genital complex, abdomen and caudal ramus) (Fig. 18).

# 1) The head (Fig. 19):

It is nearly rectangular slightly longer than wide. The antennal area is little convex and obviously protruding from the margins of the head anteriorly.

First antenna (antennule) is indistinctly segmented. Second antenna (antenna) is composed of two segments: the basal one (corpus) is large, while the terminal one is smaller and armed with a strong claw bent ventrally around the gill lamella of the fish (Fig. 19) to fix the parasite firmly to the gills of the fish host.

First maxilla (maxillul) is bilobate. The second maxilla (maxilla) is slender and two-segmented, with the proximal segment being slightly larger and unarmed while the distal segment is armed with spiniforme process. Third maxilla (maxilliped) is large and stout consisting of two segments: the proximal one is large, while the distal one is slender and armed with a terminal straight claw.

Leg 1 protopod is larger in size and conical in shape while the exopod is smaller and armed with five spines the inner spine is the largest and other is reduced in size gradually to the outward like hand fingers.

#### 2) The neck:

It is slender formed from the constriction between the 1st thoracic legs and the basis of the maxillipeds.

### 3) Trunk and urosome:

Leg 3 is large and parallel to each other. The rami are fused together to form lamella, which is folded upward along the longitudinal axis.

Leg 4 is formed of two rami, each one is a lanceolate fleshy lobe. The two lobes are fused basally at the point which is just covered by the dorsal plate (posterior margin). The endopod is slightly smaller than the exopod.

Leg 5 is a fleshy small process armed with small denticles and setae.

The genital complex is longer than wide. Caudal rami are slender and long, slightly protruding from the dorsal plate (Figure 20).

The dorsal plate is semi-circular, with ornamentation on the dorsal side and its posterior margin nearly reaches to the end of the caudal rami.

The egg sac is cylindrical, long, curved and the eggs are uniseriate.

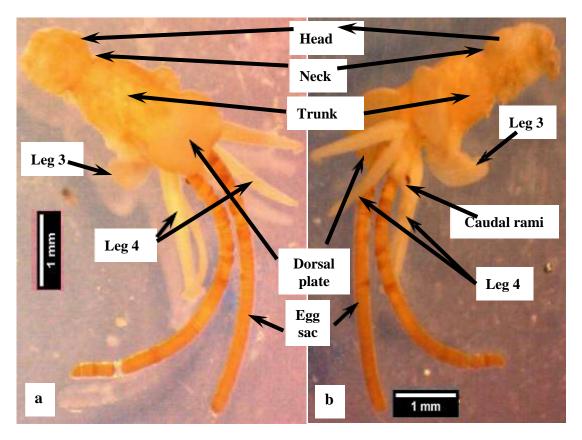


Figure 18. Light microscopic photomicrograph showing adult female Lernanthropus sarbae: a) dorsal view. b) anterolateral view



Figure 19. Light microscopic photomicrograph of *Lernanthropus sarbae* showing: a) enlarged head. b) mode of attachment of the parasite antenna to the gill lamella of the fish host.

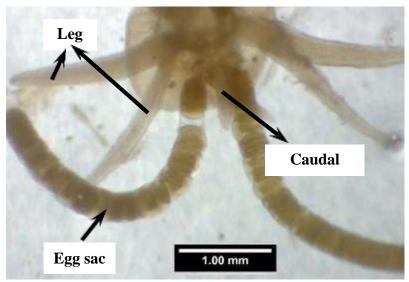


Figure 20. Light microscopic photomicrograph of *Lernanthropus sarbae* showing enlarged posterior part (ventral view).

#### **Discussion:**

Lernanthropidae is one of the large families of siphonostomatid Copepods (The 3<sup>rd</sup> largest family followed Lernaeopodidae and Caligidae), containing more than 150 species that are exclusively occurring in the gills of marine teleostes (Ho *et al.*, 2011). Lernanthropidae was originally described by Yamaguti (1963) and placed in the super family Dichelesthioidea, which includes *Lernanthropus* and other related genera of parasitic copepods. It was later revised and reported by Kabata (1979) as family Lernanthropidae (Adday, 2013).

The genus *Lernanthropus* is the oldest and largest genus of lernanthropids, having more than 105 species (Adday, 2013). Most of the species are host specific or infesting the gills of several marine teleostes inhabiting warmer waters (Al-Niaeem *et al.*, 2013). All the genera of *Lernanthropus* are characterized by having a large dorsal plate on the posterior part of the trunk and their numbers generally increase in regions of low latitudes and higher temperature (Al-Ataby *et al.*, 2012).

### Mode of attachment:

Female *Lernanthropus* use their adapted maxillipeds and antennae to firmly attach to the gill filaments of the fish host with the aid of Leg 3, which has a pair of large folded lamellae for clamping onto the gill filament of the fish (Ho *et al.*, 2011).

The mode of attachment is mentioned in detail by Khidr *et al.* (2014). They stated that the 1<sup>st</sup> and 2<sup>nd</sup> legs are small and provided with spines like hand fingers that help in parasite attachment to the adjacent gill lamellae, which increases the stability of the parasite. The 3<sup>rd</sup> and 4<sup>th</sup> legs are large and free of cuticular structure, to help in parasite position adjustment and tight attachment, but the main force for the attachment of the parasite to the fish tissue is the 2<sup>nd</sup> antenna, which is prehensile and armed with a strong claw.

In the present work the mode of attachment is very clear by light microscopy (Fig. 19), where the 2<sup>nd</sup> antenna of the parasite is showed firmly attached to the gill lamellae of the fish host. This confirms the assertion of previous work (Khidr *et al.*, 2014) that the 2<sup>nd</sup> antenna is the main force of attachment, assisted by the action of thoracic legs and maxillipeds.

#### Harmful effect:

The harmful effect of the parasite is determined by the number of parasites and mainly caused by the feeding activity and piercing action. The harmful effect of *Lernanthropus kroyeri* can be summarized as the following (Manera & Dezfuli, 2003 and Abu-Samak, 2005): 1) Desquamation, necrosis and erosion of the gill lamellae. 2) Atrophy, fibrosis and deformity of muscle bands, connective tissue and cells of the gills. 3) Haemorrhage and lumen reduction of gill blood vessels. 4) Oedema and excessive occurrence of mucous cells.

Since the intensity of infestation is very small in the present work (just one parasite in the fish host) the only noticeable pathological effect from the previously mentioned effects is the presence of excessive mucous secretion.

### Life cycle:

The life cycle of *Lernanthropus latis* is described by Brazenor and Hutson (2013) to have seven stages (2 nauplius, 3 copepodid, immature adult and adult) as following: Nauplius I is ejected from the egg sac with a clear membrane which then burst by the strong movement of the nauplius to swim freely in the water and moult to give nauplius II (6–12 hr after hatch), then the infective stage copepodid I (24–36 hr after hatch) and then copepodid II (54 hr after hatch). At this stage, the sexual dimorphism takes place and the parasite moults to give female copepodid III or male copepodid III (78 hr after hatch), then immature adult (222 hr after hatch) and then adult.

The authors also found that there was a significant interaction between salinity and temperature of the water on hatching success, with the largest hatching success at 30 °C in 35 % salinity. The parasite has a broad environmental tolerance, where the successful hatching takes place in a range of water temperatures (22–34 °C) and salinities (22–40‰).

Abdul Khalid & Shaharoum-Harrison (2014) described the life cycle of the same parasite to have 9 stages (nauplii I, nauplii II, infective copepodid, fixed copepodid I, fixed copepodid III, fixed copepodid IV, preadult, and adult), the life cycle is completed in 483 hr (20 days) under laboratory conditions, as follows: the egg expands and is ejected from the egg sac and after 15 minutes it hatches to nauplii I, which moults to nauplii II (15 hr after hatching). It is then transformed to the infective copepodid (26 - 37 hr after hatch), which swims actively, searching for the fish host, attaching to it and then moulting to the fixed copepodid I stage (72 hr after hatching), fixed copepodid II (84 hr after hatching) and then fixed copepodid IV (109 hr after hatching) where, the sexual dimorphism takes place fixed copepodid IV (109 hr after hatching), followed by moulting to preadult (207 to 483 hr after hatching) and then to the adult stage (483 hr after hatching).

## Taxonomy:

The present parasite belongs to the genus *Lernanthropus* as it has a single dorsal plate on the  $4^{th}$  thoracic somite (Adday, 2013). It resembles the widely distributed species *L. kroyeri* specially in the shape of the dorsal plate and the general structure of the body. According to Özel *et al.* (2004), the total length of the body of *L. kroyeri* is 7.5 - 15 mm which is two to four times larger than the total body length of the present specimen (3.5 - 3.6 mm).

By comparing the present parasite with previously described related species

(Table 9) the following can be concluded:

- 1) In spite that *Lernanthropus indicus* (Al-Ataby *et al.*, 2012) *Lernanthropus corniger* (Al-Niaeem *et al.*, 2013) were reported from the Arabian Gulf (Iraq), the general shape of the body, morphometric measurements and the fish host are totally different from the present parasite.
- 2) The general body structure and shape of the present parasite resemble *L.* sarbae of Olivier & Niekerk (1995) and Adday (2013).
- 3) There is a minor difference in the morphometric measurements between the present parasite and *L. sarbae* of Olivier & Niekerk (1995) and Adday (2013), this may be due to the differences in the age of the parasite studied or the technique used for measurements.
- 4) The fish host in the present study was *Acanthopagrus bifasciatus* while it was *Acanthopagrus berda* and *Rhabdosargus sarba* in Olivier & Niekerk (1995) and *A. arabicus* in Adday (2013).
- 5) The prevalence of the of the parasite in the present study was 2.2 % while it was 5.9 % in Adday (2013).
- 6) The intensity of parasite infection in the present study was 1 female while the mean was 7 in Adday (2013).

Lernanthropus sarbae (Kensley and Grindley, 1973) was first described from the gills of *Rhabdosargus sarba* from South Africa then redescribed with the aid of scanning electron microscopy from the gills of *Acanthopagrus berda* and *Rhabdosargus sarba* by Olivier & Niekerk (1995) from Lake St. Lucia, South Africa. Recently *Lernanthropus sarbae* was described from the Arabian Gulf (Iraq) from the gills of *Acanthopagrus arabicus* by Adday (2013).

For all the previous remarks the present parasite can be considered as Lernanthropus sarbae and according to the available data this is the first record of Lernanthropus sarbae in Qatar. The parasite is detected before from Acanthopagrus berda and Rhabdosargus sarba by Olivier & Niekerk (1995) and from A. arabicus by Adday (2013) but not from Acanthopagrus bifasciatus which is considered a new host for the parasite in the present study

Table 9 Comparison between female *Lernanthropus sarbae* of the present study and previously described related species (all measurements are in mm):

Character	Lernanthropus indicus	Lernanthropus corniger	Lernanthropus sarbae				
Character	Al-Ataby <i>et al.</i> (2012)	Al-Niaeem <i>et al.</i> (2013)	Olivier & Niekerk (1995)	Adday (2013)	Present work		
Shape							
Body length	46	6.9	3 - 3.5(3.3)	3.33-3.81 (3.55)	3.5 – 3.6 (3.55)		
Head	Square 13.8 ×13.7	2.30 × 1.5	Slightly longer than broad	oblong 0.88 - 1.5 (0.99) X 0.85 - 0.96 (0.91)	1.19 – 1.20 (1.19) X 0.81 – 0.96 (0.89)		
Neck	0.15 X 0.6	$0.7 \times 0.5$	-	0.19 - 0.24 (0.21) X 0.69 - 0.74 (0.72)	0.22 – 0.23 (0.22) X 0.61 – 0.69 (0.65)		
Dorsal plate	-	-	Subcircular just covering genital complex	subcircular posterior margin just covering caudal rami	subcircular with posterior margin just covering caudal rami		

<sup>\*</sup>Continued

# \*Continued

Genital complex & abdomen	12 × 24	2.3 × 3.0	-	0.50 - 0.61 (0.54) X 0.48 - 0.62 (0.54)	2.6 – 2.7 (2.65) X 0.87 – 1.1 (0.99)
Caudal ramus	long process	-	Dorsoventrally flattened	long and slender 0.46 - 0.55 (0.52) X 0.08 - 0.12 (0.10)	0.67 - 0.75 (0.71) X 0.13 - 0.16 (0.15)
Egg-sac	Long and straight	short and straight	Long and cylindrical	-	Cylindrical, long and curved
Host	Megalaspis cordyla and Carangiodes malabricus	Carangoides malabaricus and Megalaspis cordyla	Acanthopagrus berda and Rhabdosargus sarba	Acanthopagrus arabicus	Acanthopagrus bifasciatus
Prevalence	-	1.7 %, 1.4 %	-	5.9 %	2.2 %
Intensity	-	1	-	mean 7	1
Microhabitat	Gills	Gills	Gills	Gills	Gills
Locality	Arabian Gulf, Iraq	Arabian Gulf, Iraq	Lake St. Lucia, South Africa	Arabian Gulf, Iraq	Arabian Gulf, Qatar

Praniza larva of Gnathia africana Barnard, 1914

**Classification:** 

Kingdom: Animalia

Phylum: Arthropoda

Subphylum: Crustacea Brünnich, 1772

Class: Malacostraca Latreille, 1802

**Order:** Isopoda Latreille, 1781

Family: Gnathiidae Leach, 1814

Genus: Gnathia Leach, 1814

**Prevalence:** 

Three out of the 89 fish (Acanthopagrus bifasciatus) examined (3.4 %) were

found to be infested by Praniza larva of Gnathia africana. The parasite was only found

on the gills, ranging in number from one to six individuals in each fish. The parasite was

found in a mixed infection with Polylabris mamaevi and usually combined with

excessive mucous secretion on the gills (Table 10).

Two out of the 46 fish (Pomacanthus maculosus) examined (4.4 %) were found

to be infested by Praniza larva of Gnathia africana. The parasite was only found on the

gills, one individual in each fish. The parasite was found in a single infestation (Table

10).

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Table 10 Prevalence of Praniza larva of *Gnathia africana* found in the host species *Acanthopagrus bifasciatus* and *Pomacanthus maculosus* sampled from Qatar.

					Number	
Host species	No. of examine d fish	No. of infeste d fish	Prevalenc e	Microhabit at	of parasite s per fish	Mixed infestatio n
Acanthopagru s bifasciatus	89	3	3.4%	Gills	1 - 6	Polylabri s mamaevi
Pomacanthus maculosus	46	2	4.4%	Gills	1	-

### **Description Based on 5 larvae (measurements in Table 10):**

The body is long and divided into three regions (Fig. 21): head (cephalon), thorax (pereon) and abdomen (pleon):

## 1) Head (cephalon) (Fig. 22 & 23):

It is oval, broader than long (Fig. 22 & 23) and has two pairs of well-developed large compound eyes on its lateral sides. Each eye consists of many penta-shaped ocelli (Fig. 23). Cephalic appendages include: one pair of 1<sup>st</sup> antenna (antennule) which are straight, slightly shorter than the 2<sup>nd</sup> antenna and have 3 articles, with the 3<sup>rd</sup> one being the largest; 2<sup>nd</sup> antenna (antenna) are straight, longer than 1<sup>st</sup> antenna and have 4 articles, with the 4<sup>th</sup> one being the largest; mandibles (Fig. 23c) which are knife-shaped and armed with 9 teeth (two are small on the terminal end and 7 are larger triangular in shape and directed to the backward); 1<sup>st</sup> maxilla (maxillule); 2<sup>nd</sup> maxilla (maxilla) and; 3<sup>rd</sup> maxilla (maxilliped), where coupling hooks are abscent and the 1<sup>st</sup> article has 6 small

teeth.

#### 2) Thorax (pereon) (Fig. 21 & 22):

It is nearly twice as long as wide and composed of 7 segments (pereonites), with the 1<sup>st</sup> one being fused to the head while the remaining are free (Fig. 22). The anterior margin of pereonite 1 is convex and much wider than the posterior margin of the head. Pereonite 1 is slightly narrower than pereonite 2 and pereonite 3 is wider than the preceding ones. The 1<sup>st</sup> three pereonites are distinctly segmented, while pereonites 4, 5 and 6 are not segmented and fused with each other to form the largest, elongated part of the body which is swollen when the larva takes its blood meal. Pereonite 7 is small, visible dorsally, with its posterior end rounded and overlapping with the first abdominal segment (pleonite 1). There are five pairs of thoracic legs (pereopods), each with 6-segments, covered with setae and provided with prominent, sharp and posterior-pointing spine on its terminal end.

### 3) The abdomen (Pleon) (Fig. 24):

Is composed of six segments (pleonites), which are narrower than the pereonites and each carrying a pair of legs (pleopods). The 1<sup>st</sup> five pleonites are free while the 6th pleonite is fused with the telson to form the pleotelson. Pleopods are biramous, foliate and have long simple plumose setae. The pleotelson is triangular, with straight lateral margins, fringed with setae in its terminal part and pears the uropods, which have long endopods reach to the end of the pleotelson and shorter expods, both of which are fringed with many long plumose setae.

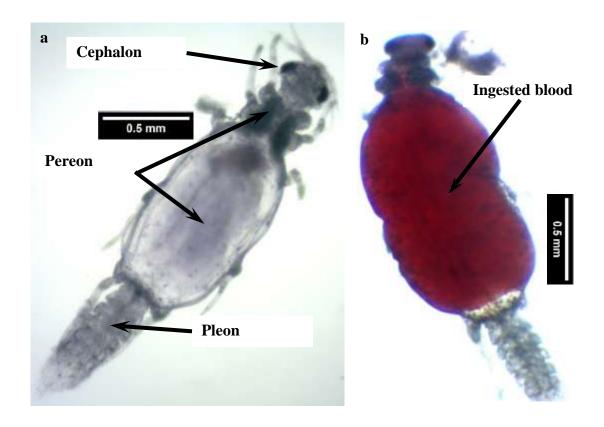


Figure 21. Light microscopic photomicrograph showing Praniza larva of *Gnathia africana*: a) dorsal view. b) dorsal view after finishing a blood meal.

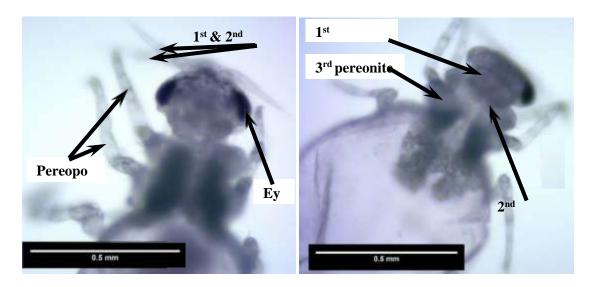


Figure 22 Light microscopic photomicrograph showing enlarged anterior portion of Praniza larva of *Gnathia africana*.

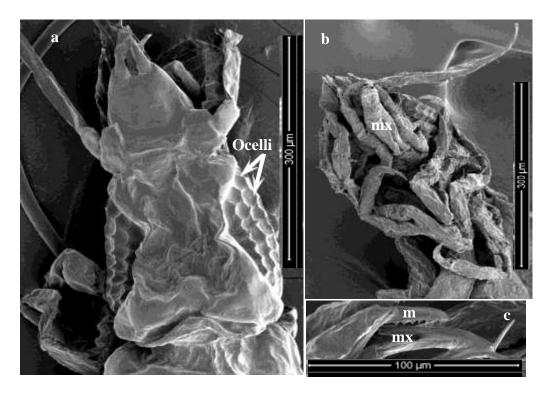


Figure 23. SEM of praniza larva of *Gnathia africana* showing enlarged cephalon a) dorsal view showing the penta-shaped ocelli of the well-developed eyes. b) ventral view. c) enlarged mandible (m) and maxilliped (mx).

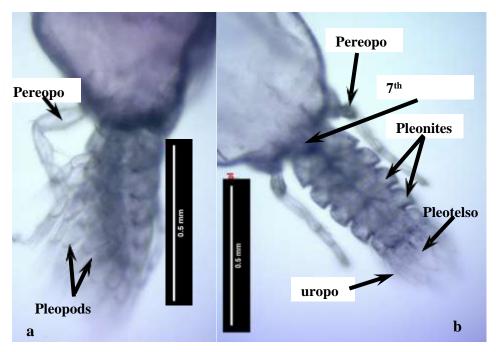


Figure 24. Light microscopic photomicrograph showing enlarged posterior portion of Praniza larva of *Gnathia africana*: a) lateral view. b) dorsal view.

#### **Discussion:**

Isopods are one of the crustacean groups (class Malacostraca) that can live in aquatic or terrestrial habitats, as free living (filter feeder or marine scavenger) or parasitic, in one or more stage of their life cycle. Their size varies from microscopic to 16-inch-long, with highly variable shapes (Adday, 2013).

#### Harmful effect:

The harmful effects of praniza larva on the fish host include necrosis, fusion of gill lamellae and mechanical damage to gill epithelium. In case of severe infestation, it can cause hypoxia as a result of total loss of the gill function. A single parasite can ingest up to 1.89 mg of blood and this consider a large amount knowing that the total amount of blood in a fish is 1/20 of its weight. Accordingly, severe infestations can cause severe anemia that can lead to death, particularly in small fish (Marino *et al.*, 2004). Furthermore, praniza larva can act as a vector in transmitting *Haemogregarina bigemina* which is a protozoan blood parasite for fish (Smit, 2002)

### Mode of attachment:

Isopods use their anterior pereopods and mouth parts to attach firmly to the host fish and the appendages which used in attachment usually armed with curved terminal spines.

#### Life cycle:

As described by Smit & Basson (2003) the life cycle of *G. Africana* contains seven stages: 3 unfed zuphea larval stages and 3 praniza larval stages, plus the adult. The full cycle takes about 62 days to complete, as follows:

The eggs hatch to give stage 1 zuphea larva which immediately search about the fish host, attach to it, feed on lymph fluid or blood for about two hours (some fish can remove the larvae from their body and may eat it) and then transforms to praniza 1

phase.

After taking its blood meal praniza larva leave the fish host and hide in sponges or tunicates, where it moults after 8 days to give stage zuphea-2 larvae which after two days fed again on a fish host (from 2 to 3 hours) and moult to give praniza-2 larvae then leave the fish host again to the sponge.

After 10 days, it moults to give zuphea 3 larvae which fed again (from 3 to 23 hours) and moult to give praniza 3 larvae (sexual dimorphism takes place in this stage) which moult to give adult male 10 days after feeding and adult female 17 days after feeding.

Fertilization of the egg usually occurs within 24 hours after female moulting. Development of the young larvae and their release follows and the life cycle takes place continuously, not following any seasonal patterns.

## Taxonomy:

The Gnathiidae family is characterized by the presence of only 5 pairs of pereopods (walking legs) instead of the 7 pairs that characterize all other isopods. The taxonomy of this family is notoriously difficult because the same species have different stages in their life cycle, with many morphological differences that lead early taxonomists to miss the link between larvae, female and male and to classify them as different species, from different genera, until later the genus *Gnathia* (Leach, 1814) was erected (Smit, 2002).

Gnathia africana was first described by Barnard (1914) from Southern Africa. Later on, Smit et al. (1999) redescribed the same parasite from the same location. In the same location Smit & Basson (2002) described a new species (Gnathia pantherine) from the gills, nares and buccal cavity of Poroderma pantherinumome and some elasmobranch species, but the authors considered the presence of the parasite in the

caudal and pelvic fins as a sampling artifact.

Since the taxonomy of gnathiids isopod is based solely on the morphology of the free-living adult male (Cohen and Poore, 1994), making identification of females and parasitic larvae difficult (Smit and Davies, 2004). However, Smit & Basson (2002) clarified that the praniza larva of *G. africana* and *G. pantherina* are very similar in the basic morphology, but, the two species can be distinguished by the following:

- 1) The pleotelson of *G. Africana* larva has straight anterio-lateral margins comparing to *G. pantherine* which has concave anterio-lateral margins.
- 2) The mandible of *G. Africana* larva is armed with 9 -10 teeth comparing to *G. pantherine* which have only 8 teeth.
- 3) The coupling hooks are absent in the maxilliped of *G. Africana* larva, which has 5 7 teeth on its terminal part comparing to *G. pantherine* larva where the coupling hooks are present in the maxilliped and which has 3 5 on its terminal part.
- 4) G. Africana larva is much smaller than G. pantherine larva.

Adday (2013) recorded the first occurrence of praniza larva in fishes of the Arabian gulf (Iraq) from the gills of some telosti & elasmobranch species, but the details of the mandible and maxilliped were not mentioned, so he only reached to the genus level and did not mention the species of the larva. In the same year, Bayoumy *et al.* recorded the presence of praniza larva of *G. pantherine* in the Arabian Gulf (Saudi Arabia, Damam) from the gills, pectoral fins and mouth cavity of *Epinephelus tauvina*. These authors confirmed the presence of the parasite in the caudal and pelvic fins of the fish and that is not a sampling artifact, as mentioned by Smit & Basson (2002).

By comparing the present praniza larva with the previously described related species (Table 11) the following can be noted:

- 1) Body length of the present parasite in the range of G. *Africana* Smit *et al.* (1999) and smaller than all other related species: *G. pantherine* (Smit & Basson, 2002 and Bayoumy *et al.*, 2013) and *Gnathia* sp. Adday (2013).
- 2) The mandible of the present parasite is armed with 9 teeth, which is in the range of G. *Africana* Smit *et al.* (1999) and *G. pantherine* Bayoumy *et al.* (2013), while *G. pantherine* Smit & Basson (2002) is armed with only 8 teeth.
- 3) The coupling hook of the maxilliped is absent in the present parasite, which is like *G. Africana* Smit *et al.* (1999) and *G. pantherine* Bayoumy *et al.* (2013), while it is present in *G. pantherine* Smit & Basson (2002).
- 4) The 1<sup>st</sup> article of maxilliped of the present parasite is armed with 6 teeth, which is in the range of G. *Africana* Smit *et al.* (1999), while *G. pantherine* Smit & Basson (2002) and *G. pantherine* Bayoumy *et al.* (2013) is armed with only 3 5 teeth
- 5) The pereon of the present parasite is twice as long as wide, which is like G. *Africana* Smit *et al.* (1999) and *G. pantherine* Smit & Basson (2002), while it is one and half times as long as wide in *G. pantherine* Bayoumy *et al.* (2013).
- 6) Pereonite 7 is dorsally visible and overlapping first pleonite in the present parasite, which is like *G. Africana* Smit *et al.* (1999) and *G. pantherine* Smit & Basson (2002), while it is not visible in Gnathia sp. Adday (2013).
- 7) Pleotelson anterio-lateral margins is straight in the present parasite, which is like G. *Africana* Smit *et al.* (1999) and *G. pantherine* Bayoumy *et al.* (2013), while it is concave in *G. pantherine* Smit & Basson (2002) and *Gnathia* sp. Adday (2013) (from the drawing, not mentioned in the text).

- 8) Acanthopagrus bifaciates & Pomacanthus maculosus are the fish host of the present parasite while it was some some telosti & elasmobranch species (not including Acanthopagrus bifaciates & Pomacanthus maculosus) in all the previously described related species (Smit et al. (1999), Smit & Basson (2002) Adday (2013) and Bayoumy et al. (2013)).
- 9) The prevalence of the parasite in the present study was 3.4 % in *Acanthopagrus bifaciates* and 4.4 % in *Pomacanthus maculosus*, which is substantially less than the prevalence of the parasite in the previous studies (69 % in Adday (2013) and 58.33% in Bayoumy *et al.* (2013).
- 10) The intensity of parasite infestation in the present study was 1 6 in *Acanthopagrus bifaciates* and 1 in *Pomacanthus maculosus* and this much smaller than the intensity of parasite infestation in the previous studies (mean 12.3 in Adday (2013) and 8 -22 in Bayoumy *et al.* (2013)).
- 11) The location of the present parasite is Qatar (Arabian Gulf), while it was Southern Africa in *G. Africana* Smit *et al.* (1999) and *G. pantherine* Smit & Basson (2002), while it was Iraq (Arabian Gulf) in *Gnathia* sp. Adday (2013) and Saudi Arabia (Arabian Gulf) in Bayoumy *et al.* (2013).

From the above-mentioned comparison and discussion, the following can be concluded:

the present parasite is more similar to *G. Africana* Smit *et al.* (1999) as it is smaller in size, the mandible is armed with 9 teeth, the maxilliped has 6 teeth on its 1<sup>st</sup> article, the coupling hook is absent and the antero-lateral margins of the pleotelson is straight (as previously mentioned all these characters were used by Smit & Basson (2002) to differentiate between the praniza larva of *G. Africana* and *G. pantherine*).

Gnathia sp. Adday (2013) is more similar to G. pantherine Smit & Basson (2002) as it is larger in size and the antero-lateral margins of the pleotelson is concave,

but the details of the mandible and maxilliped is missing which make it difficult to confirm the identification of the species.

G. pantherine Bayoumy et al. (2013) has some common characters with the pranazia larva of G. Africana Smit et al. (1999) (the coupling hook is absent and the pleotelson is straight). At the same time, it has some common characters with the pranazia larva of G. pantherine Smit & Basson (2002) (it is larger in size, the maxilliped has 3 - 5 teeth on its 1<sup>st</sup> article). In addition, it has some characters that differs from both of them (the mandible is armed with 7 -9 teeth while it is only 8 in G. pantherine Smit & Basson (2002) and 8 - 10 in G. Africana Smit et al. (1999) and the pereon is one and half times as long as wide while it is twice as long as wide in both G. pantherine Smit & Basson (2002) and G. Africana Smit et al. (1999)). From the previous notes G. pantherine Bayoumy et al. (2013) may be considered as another species, differing from both G. Africana Smit et al. (1999) and G. pantherine Smit & Basson (2002).

According to the available data, this study is the first record of the pranazia larva of G. *Africana* from the Arabian Gulf (Qatar). More over *Acanthopagrus bifaciates* & *Pomacanthus maculosus* are considered new hosts for this parasite.

Table 11 Comparison between Praniza larva of *Gnathia africana* of the present study and previously described related species (all \*Continued \*

*Continued	,					
	Gnathia sp.	Gnathia pant	herina	Gr	Gnathia africana	
Character	Adday (2013)	Smit & Basson (2002)	Bayoumy <i>et al.</i> (2013)	Smit et al. (1999)	Present work	
Shape			A.Zama	12	3.5 mm	
Body length	3.54 - 4.69 mm (4.5 mm)	3.9 - 5.8 mm	3.8 - 5.9 mm	1.1 - 3.9 mm	2.38 – 3.27 (2.87) mm	

<sup>\*</sup>Continued

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·	Gnathia sp.	Gnathia pani	therina	Gnathia africana		
Character	Adday (2013)	Smit & Basson (2002)	Bayoumy <i>et al</i> . (2013)	Smit et al. (1999)	Present work	
Head ventral view	-			34		
Mandible	-	Only 8 teeth	7-9 teeth	8-10 teeth	9 teeth	
Maxilliped coupling hooks	-	Present	Abscent	Abscent	Abscent	
Teeth on maxilliped	-	3 -5	3 -5	5 - 7	6	
Pereon	-	Twice as long as wide	One and half times as long as wide	Twice as long as wide	Twice as long as wide	
Pereonite 7	Not visible	Dorsally visible, overlapping first pleonite	-	Dorsally visible, overlapping first pleonite	Dorsally visible, overlapping first pleonite	

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	Gnathia sp.	Gnathia pant	herina	Gr	nathia africana
Character	Adday (2013)	Smit & Basson (2002)	Bayoumy <i>et al</i> . (2013)	Smit et al. (1999)	Present work
Pleon & pleotelson					Constant of the second
Telson & uropod	Ur→		-		
Pleotelson anterio- lateral margins	Concave (from the drawing not mentioned in the text)	Concave	Straight	Straight	Straight
*Continued  Host	Chiloscyllum arabicum, some telosti & elasmobranch species	Poroderma  pantherinumome & some elasmobranch species	Epinephelus tauvina	-	Acanthopagrus bifaciates & Pomacanthus maculosus
Prevalence	69 %,	-	58.33%	-	3.4 %, 4.4 %
Intensity	mean 12.3	-	8 - 22	-	1 – 6 & 1

# \*Continued

	Gnathia sp.	Gnathia pant	Gnathia pantherina		thia africana
Character	Adday (2013)	Smit & Basson (2002)	Bayoumy <i>et al</i> . (2013)	Smit et al. (1999)	Present work
Microhabitat	Gills	Gills, nares and buccal cavity	Gills, pectoral fins and mouth cavity	-	Gills
Locality	Arabian Gulf, Iraq	Southern Africa	Arabian Gulf, Saudi Arabia	Southern Africa	Arabian Gulf, Qatar

Polylabris mamaevi Ogawa & Egusa, 1980.

**Classification:** 

**Kingdom:** Animalia

**Phylum:** Platyhelminthes Minot, 1876

Subphylum: Neodermata Ehlers, 1985

Class: Monogenoidea Bychowsky, 1937

Subclass: Heteronchoinea Boeger et Kritsky, 2001

Order: Mazocraeidea Bychowsky, 1937

Family: Microcotylidae

Genus: Polylabris Euzet et Cauwet, 1967

**Prevalence:** 

68 out of the 89 fish (Acanthopagrus bifasciatus) examined (76.4%) were found

to be infested by *Polylabris mamaevi*. The parasite was only found on the gills, ranging

in number from one to more than 50 individuals in each fish. The parasite was

sometimes found in a mixed infection with Alella sp., Caligus haemulonis,

Lernanthropus sarbae or Pranzia larva of Gnathia Africana and usually combined with

excessive mucous secretion on the gills. Pale liver and gills and injuries in front of the

fish are usually combined with heavily parasitic infection (Table 12).

Two out of the 46 fish (*Pomacanthus maculosus*) examined (4.4 %) were found

to be infested by *Polylabris mamaevi*. The parasite was only found on the gills, one

individual in each fish. The parasite was found in a single infestation (Table 12). Figures

(from 25 to 29) showing all the details of the parasite.

80

Table 12 Prevalence of *Polylabris mamaevi* (Ogawa & Egusa, 1980) found in the host species *Acanthopagrus bifasciatus* and *Pomacanthus maculosus* sampled from Qatar.

Host species	No. of examine d fish	No. of infeste d fish	Prevalenc e	Microhabit at	Numbe r of parasite s per fish	Mixed infestation
Acanthopagr us bifasciatus	89	68	76.4%	Gills	1 – 50+	Alella sp., Caligus haemulonis, Lernanthrop us sarbae or Pranzia larva of Gnathia Africana
Pomacanthus maculosus	46	2	4.4%	Gills	1	-

## **Discussion:**

A comparison between *Polylabris mamaevi* of the present study and previously described related species is givin in Table 13. According to the available data this is the first record of the parasite from the Arabian Gulf (Qatar). Moreover, *Acanthopagrus bifasciatus* and *Pomacanthus maculosus* are considered new fish hosts for the parasite



Figure 25. Light microscopic photomicrograph of Polylabris mamaevi ventral view



Figure 26. Light microscopic photomicrograph of *Polylabris mamaevi* ventral view showing enlarged copulatory organ

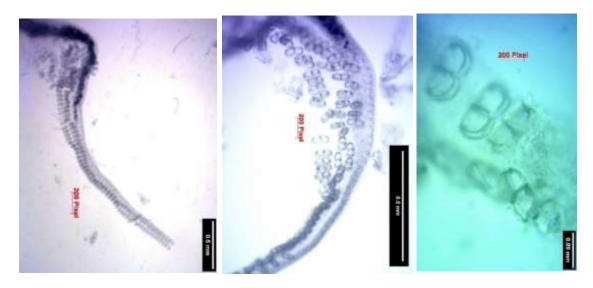


Figure 27 Light microscopic photomicrograph of *Polylabris mamaevi* ventral view showing enlarged haptor

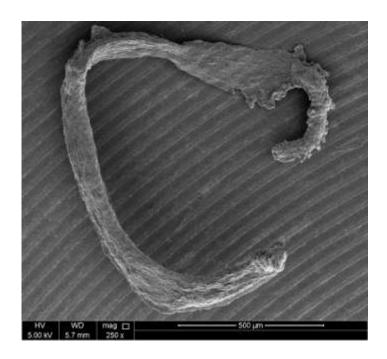


Figure 28. SEM of Polylabris mamaevi dorsal view.

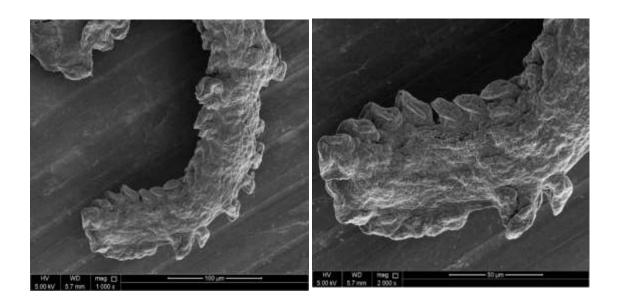


Figure 29 SEM of *Polylabris mamaevi* dorsal view showing enlarged posterior end

Table 13 Comparison between Polylabris mamaevi of the present study and previously described related species (all measurements are in

\*Continued P. tubicirrus P. lingaoensis P. mamaevi Parasite Santos et al., Tingbao et al., Bayoumy et al., Tingbao *et al.*, (2007) Present study (1996)(2007)(2015) Figure 1.356 1.231 3.7 2.8 (1.837 - 4.122) 3.4 (1.98 - 4.74) length Width at level of transverse portion 0.446 (0.359 - 0.595)0.28 (0.2 - 0.41) of germarium

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<sup>\*</sup>Continued

# \*Continued

	P. tubicirrus P. lingaoensis			P. mamaevi		
Parasite	Santos <i>et al.</i> , (1996)	Tingbao <i>et al.</i> , (2007)	Bayoumy <i>et al.</i> , (2015)	Tingbao <i>et al.</i> , (2007)	Present study	
			<b></b>	Paired elliptical 0.061	Paired elliptical 0.08	
prohaptoral sucker	Paired;	Paired elliptical to	Paired elliptical to	(0.049 – 0.074) X 0.057	(0.07 - 0.10) X 0.06	
		subcircular; subcircular;		(0.049 - 0.072)	(0.05 - 0.07)	
				spherical, 0.043 (0.032–		
Pharynx	-	-	-	0.051)	0.043 (0.04 - 0.05)	
Level of intestinal		At the level of	At the level of	At the level of male	Above the male	
bifurcation		common genital pore	common genital	copulatory organ	copulatory organ	
Haptor	5	Beginning at the	Beginning at the	Beginning at the level of	Beginning at the level of	
	Reaches the	level of posterior	level of posterior	posterior testes 0.446	posterior testes 1.3 (0.65	
	testes level	testes	testes	(0.359 - 0.595) long	-1.72)	

<sup>\*</sup>Continued

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	P. tubicirrus	P. lingaoensis		P. mamaevi		
Parasite	Santos <i>et al.</i> , (1996)	Tingbao <i>et al.</i> , (2007)	Bayoumy <i>et al.</i> , (2015)	Tingbao <i>et al.</i> , (2007)	Present study	
Clamps shape	-	3		5	32	
,				Parallel subequal rows of	Parallel subequal rows	
clamps	-	-	-	27 – 47	62 (51-74)	
anterior clamp				$0.058 \ (0.051 - 0.068)$	0.04 (0.04 0.00)	
	-	-	-	wide	0.06 (0.04-0.08)	
Common genital						
pore	-	-	midventral	midventral	midventral	
Genital atrium	-	-	-	unarmed	unarmed	

<sup>\*</sup>Continued

	P. tubicirrus	P. lingaoensis		P. mamaevi		
Parasite	Santos <i>et al.</i> , (1996)	Tingbao <i>et al.</i> , (2007)	Bayoumy <i>et al.</i> , (2015)	Tingbao <i>et al.</i> , (2007)	Present study	
		5 to 7 testes	6 to 8 testes			
Testes		intercaecal in	intercaecal in	9 to 14 testes intercaecal	13 (12-16) intercaecal in	
Testes	-	posterior half of	posterior half of	in posterior half of trunk	posterior half of trunk	
		body	body			
				conical comprising inner		
		conical comprising		tube and outer sheath	Bear-shaped comprising	
copulatory organ	-	inner tube and outer	-	0.053 (0.046 – 0.060) X	inner tube and outer	
		sheath		0.033 (0.027 –	sheath 0.05 X 0.04	
				0.037)		
		pretesticular,	pretesticular,			
Germarium	-	intercaecal	intercaecal	pretesticular, intercaecal	pretesticular, intercaecal	
Vaginae	-	unarmed	unarmed	unarmed	unarmed	

<sup>\*</sup>Continued

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	P. tubicirrus P. lingaoensis		aoensis	P. mamaevi			
Parasite	Santos <i>et al.</i> , (1996)	Tingbao <i>et al.</i> , (2007)	Bayoumy <i>et al.</i> , (2015)	Tingbao <i>et al.</i> , (2007)	Present study		
	Diplodus	Ambassis	Acanthopagrus		Acanthopagrus		
Host	Dipiodus	Amoussis		Siganus fuscescens	bifasciatus,		
	argenteus	gymnocephalus,	bifasciatus		Pomacanthus maculos		
Prevalence	-	-	53.3%	-	76.4%, 4.4%		
Intensity	-	-	-	-	1 - 50+, 1		
Microhabitat	Gills	Gills	Gills	Gills	Gills		
	Rio de						
Locality	Janeiro	Gulf of Tonkin	The Red Sea,	Gulf of Tonkin (South	Anchion Culf (Oct		
	coast	(South China Sea)	Hurghada (Egypt)	China Sea)	Arabian Gulf (Qatar		
	(Brazil)						

# Stephanostomum sp.

### **Prevalence:**

one out of the 46 fish (*Pomacanthus maculosus*) examined (2.1 %) was found to be infested by *Stephanostomum* sp. One individual parasite was only found in the intestine of one fish. The parasite was found in a double infestation with *Cucullanus* sp. (Table 14). Figures (from 30 to 32) showing all the details of the parasite.

Table 14 Prevalence of Stephanostomum sp. found in the host species Pomacanthus maculosus sampled from Qatar

	No. of	No. of			Number	
	- 100	_,,,,			of	Mixed
Host species	examined	infested	Prevalence	Microhabitat	parasites	infestation
	fish	fish			r	
					per fish	
Pomacanthus						Cucullanus
maculosus	46	1	2.1%	intestine	1	sp.



Figure 30. Light microscopic photomicrograph of Stephanostomum sp. ventral view

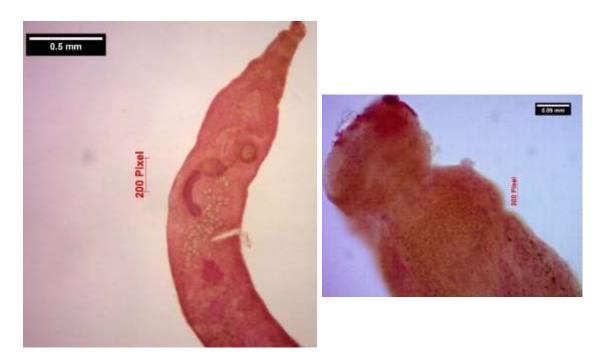


Figure 31. Light microscopic photomicrograph of Stephanostomum sp. ventral view showing enlarged anterior end.

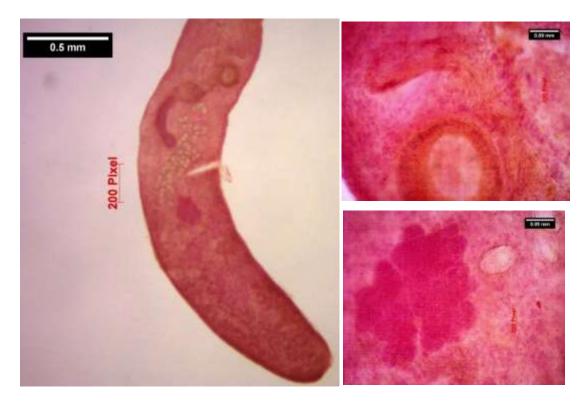


Figure 32. Light microscopic photomicrograph of *Stephanostomum* sp. ventral view Showing enlarged posterior end

# Cucullanus sp.

## **Prevalence:**

Seven out of the 46 fish (*Pomacanthus maculosus*) examined (15.2 %) was found to be infested by *Cucullanus* sp. The parasite was only found in the intestine, ranging in number from one to eight individuals in each fish. The parasite was once found in a double infestation with *Stephanostomum* sp. (Table 15). Figures 33, 34 and 36 showing the details of the parasite.

According to the available data this is the first record of *Cucullanus* sp. from the fish of Qatari water.

Table 15 Prevalence of Cucullanus sp found in the host species Pomacanthus maculosus sampled from Qatar

					Numbe	
Host species	No. of examine d fish	No. of infeste d fish	Prevalen ce	Microhabit at	r of parasit es per fish	Mixed infestation
Pomacanth us maculosus	46	7	15.2%	intestine	1-8	Stephanostom um sp.

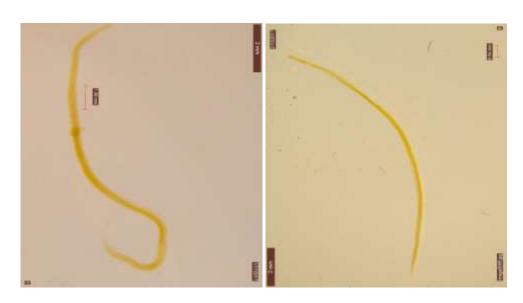


Figure 33. Light microscopic photomicrograph showing lateral view of male and female Cucullanus sp.

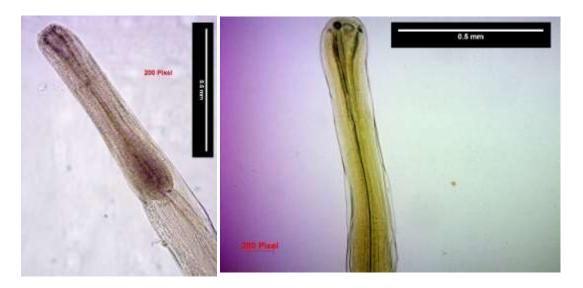


Figure 34. Light microscopic photomicrograph showing enlarged anterior end of *Cucullanus* sp.



Figure 35. Light microscopic photomicrograph showing enlarged posterior end of *Cucullanus* sp.

# Dujardinascaris sp.

### **Prevalence:**

Two out of the 89 fish (*Acanthopagrus bifasciatus*) examined (2.2 %) were found to be infected with *Dujardinascaris* sp. Three nematodes were found alone in the intestine of one fish and 2 encysted parasites were found on the testes of another fish. (Table 16). Figures 36 and 37 showing the details of the parasite.

According to the available data this is the first record of *Dujardinascaris* sp. from the fish of Qatari water.

Table 16 Prevalence of Dujardinascaris sp. found in the host species Pomacanthus maculosus sampled from Qatar

	No. of	No. of			Number of	Mixed	
Host species	examine	infeste	Prevalenc e	Microhabita t	parasite	infestatio	
	d fish	d fish	Č	·	s per	n	
					fish		
Acanthopagru	0.0			Intestine			
s bifasciatus	89	2	2.2%	and testes	1-8	-	



Figure 36. Light microscopic photomicrograph showing enlarged anterior end of Dujardinascaris sp.



Figure 37. Light microscopic photomicrograph showing enlarged posterior end of *Dujardinascaris* sp.

#### **CHAPTER 3: FISH HOSTS**

During the present study, two fish species associated to Qatar coral reefs were examined for health abnormalities, including parasitic infestations.

The two-fish species are:

- 1) Acanthopagrus bifasciatus (Two-bar seabream): 89 fish were examined, the length of the fish ranges from 18.5 to 35.4 cm while the weight ranges from 114 to 597 g.
- 2) *Pomacanthus maculosus* (Yellow-bar angelfish): 35 fish were examined, the length of the fish ranges from 14 30 cm while the weight ranges from 50.7 760 g

Anatomical observations and parasitic infections are reported separately in the following sections.

### 1- Anatomical observations

During the present study, several external and internal features appeared in some of the studied fish specimen. These features were considered abnormal by comparison with the other examined fish from the same batch (having the same conditions regarding fishing method, handling and degree of freshness). Two types of anatomical observations were considered: 1- those appearing combined with particular parasitic infection and 2- those appearing regardless of the presence of parasitic infection.

### Acanthopagrus bifasciatus:

Anatomical observations combined with parasitic infection:

- Excessive mucous secretion on gills and red dark filaments of gills, usually combined to parasitic infection with Monogenea and/or Copepoda.
- Pale liver and gills and injuries in front of the fish are usually combined with

- heavily parasitic infection with Monogenea (Fig. 38).
- Abnormal branches in the gills, usually combined with infection by the copepod *Caligus haemulonis*.

## Anatomical observations not combined with parasitic infection:

- In only one fish an unknown balloon-like structure was found in the gill chamber of the fish (Fig. 39).
- Green gall bladder (Fig. 40) and fat deposits surrounding the intestine, broken frontal teeth.



Figure 38. Images of different parts (same fish) of the fish host Acanthopagrus bifasciatus sampled from Qatar infested by more than 50 worms of the monogenean parasite Polylabris mamaevi: a) frontal view of the fish showing many injuries in the frontal side of the head. b) the worms encountered from the fish. c) Pale gills.



Figure 39. Images of an unknown structure found in the fish host Acanthopagrus bifasciatus sampled from Qatar: a) zoom out of the gill chamber of the fish showing the structure. b) zoom in of the gill chamber of the fish showing the structure after removal from the fish.

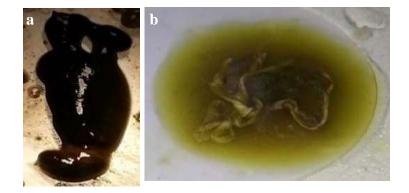


Figure 40. Images of the gall bladder found in the fish host Acanthopagrus bifasciatus sampled from Qatar with dark green color: a) the gall bladder after removal from the fish. b) the gall bladder after opening showing the green bile.

### Pomacanthus maculosus:

All the anatomical observations noticed were not combined with parasitic infection and included: Red dots on the gills, patches of the skin without scales (Fig. 41) and the presence of different percentages of black sections in the liver ranging from just black edges to totally black liver (Fig. 42).

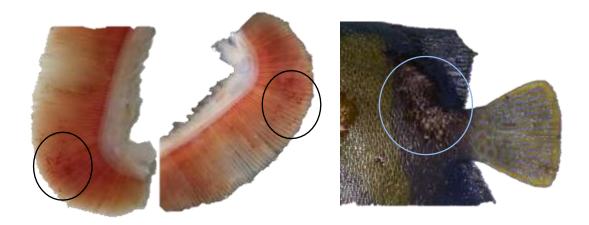


Figure 41. Images of different parts of the fish host *Pomacanthus maculosus* sampled from Qatar: a) Enlarged part of the gills showing red dots on gill lamellae (black circles). b) Enlarged part of the posterior end showing a patch of the skin without scales (blue circle).



Figure 42. Images of liver from different fish *Pomacanthus maculosus* sampled from Qatar showing different percentages of black parts in the liver ranging from just black edges to totally black liver.

# 2- Intensive parasitic infections

The fishes with two species of parasites or with four parasites or more per one fish were considered to have intensive parasitic infestation which is a health abnormality (Moore, *et al.*, 1997).

For *Acanthopagrus bifasciatus* 33 (37.1%) fish out of 89 were intensively infested by parasites, while for *Pomacanthus maculosus* 3 (6.5%) fish out of 46 were intensively infested by parasites.

## 3- Prevalence of the parasites in fish hosts

Eight different parasitic species were found infecting the two studied fish species. Table 17 and 18 summarizes the parasites encountered during the present study from each of the two fish species, their groups, microhabitat, Prevalence and intensity.

Table 17 Parasites encountered during the present study from *Acanthopagrus bifasciatus*, their group, host, microhabitat, prevalence and intensity.

Parasite group	Parasite species	Microhabitat	Prevalence	Intensity
Monogenea	Polylabris mamaevi	Gills	76.4 %	1 – 50+
C1-	Caligus haemulonis	Gills	5.6 %	1 - 3
Copepoda	Alella sp.	Gills	4.5 %	1 - 2
	Lernanthropus sarbae	Gills	2.2 %	1
Isopoda	Gnathia africana	Gills	3.4 %	1 - 6
NT 1	Dujardinascaris sp.	Intestine	2.2 %	2 - 3
Nematoda	Cucullanus sp.	Intestine	15.2 %	1 - 8

Table 18 Parasites encountered during the present study from *Pomacanthus maculosus*, their group, host, microhabitat, prevalence and intensity.

Parasite group	Parasite species	Microhabitat	Prevalence	Intensity
Monogenea	Polylabris mamaevi	Gills	4.4 %	1
Trematoda	Stephanostomum sp.	Intestine	2.1 %	1
Isopoda	Gnathia africana	Gills	4.4%	1
Nematoda	Cucullanus sp.	Intestine	15.2 %	1 - 8

Out of the 135-fishes, examined in this study,84 (62.2 %) were found infected with one or more of the following groups of parasites (in descending order): Monogenea 70 (51.9 %), Copepoda 11(8.5 %), Nematoda 9 (6.6 %), Isopoda 5 (3.7%), and

Trematoda 1 (0.8%).

## Acanthopagrus bifasciatus:

Out of the examined 89-fishes 72 (80.9 %) were found infested with one or more of the following groups of parasites (in descending order): Monogenea 68 (76.4 %), Copepoda 11 (12.4 %), Isopoda 3 (3.4%) and Nematoda 2 (2.2 %) (Table 19 and Fig. 43). The prevalence of each parasite species is mentioned in detail in Table 17.

# Pomacanthus maculosus:

Out of the examined 46-fishes 12 (26.1 %) were found infected with one or more of the following groups of parasites (in descending order): Nematoda 7 (15.2%), Monogenea 2 (4.4 %), Trematoda 1 (2.1 %) and Isopoda 2 (4.4 %) (Table 19 and Fig. 43). The prevalence of each parasite species is mentioned in detail in Table 18.

Table 19 The prevalence of parasite groups in fish hosts sampled from Qatar.

	No of	E	ish	Parasites										
Fish	No. of			Mon	ogen	Coj	pepo	Nei	mato	Iso	pod	Tren	nato	
species	fish examin	ШЕ	infested		ea		da		da		a		da	
species	ed	N	%	No	%	N o.	%	N o.	%	N o.	%	No	%	
Acanthopa grus bifasciatus	89	72	80. 9	68	76. 4	11	12. 4	2	2.2	3	3. 4	0	0	
Pomacanth us maculosus	46	12	26. 1	2	4.4	0	0	7	15. 2	2	4. 4	1	2.1	
Total	135	84	62. 2	70	51. 9	11	8.5	9	6.6	5	<ul><li>3.</li><li>7</li></ul>	1	0.8	

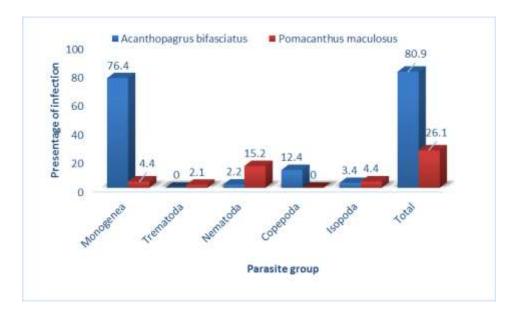


Figure 43. Prevalence of different parasite groups in the examined fishes.

### 4- Microhabitat of the parasites

Parasite species were found in 4 different microhabitats within the two considered fish species: gills, intestine, stomach and testes. Table 20 shows the microhabitat of each parasite species and its percentage regardless of the host fish. Nevertheless, both fish hosts exhibited dissimilar patterns of infestation; a general description of the infestation patterns for each fish host is given here below.

Acanthopagrus bifasciatus: The common microhabitat for Monogenea,
Copepoda and Isopoda was the gills (80.2 %) while the common microhabitats for
Nematoda were the intestine (1.1 %) and testes (1.1 %) (Table 21 and Figure 44). The
microhabitat of each parasite species is mentioned in detail in Table 20.

<u>Pomacanthus maculosus:</u> The common microhabitat for Nematoda and Trematoda was the intestine (14.6 %) and the common microhabitat for Monogenea

and Isopoda was the gills (8.5 %) (Table 21 and Figure 44). The microhabitat of each parasite species is mentioned in detail in table 20.

Table 20 The microhabitat of each parasite species in fish hosts sampled from Qatar.

Parasita anacias	Gills	Intestine	Stomach		Testes			
Parasite species	No.	%	No.	%	No.	%	No.	%
Polylabris mamaevi	68	100	0	0	0	0	0	0
Caligus haemulonis	s haemulonis 5 100		0	0	0	0	0	0
Alella sp.	4	100	0	0	0	0	0	0
Lernanthropus sarbae	2	100	0	0	0	0	0	0
Gnathia africana	3	100	0	0	0	0	0	0
Dujardinascaris sp.	0	0	1	50	0	0	1	50
Cucullanus sp.	0	0	6	85.7	1	14.3	0	0

Table 21 The microhabitat of parasite groups in fish hosts sampled from Qatar

Fish species	G	ills	Inte	stine	Stomach		Tes	tes
rish species	No.	%	No.	%	No.	%	No.	%
Acanthopagrus bifasciatus	70	97	1	1.4	0	0	1	1.4
Pomacanthus maculosus	4	8.5	7	15.2	1	2.1	0	0
Total	74	54.8	8	5.9	1	0.7	1	0.7

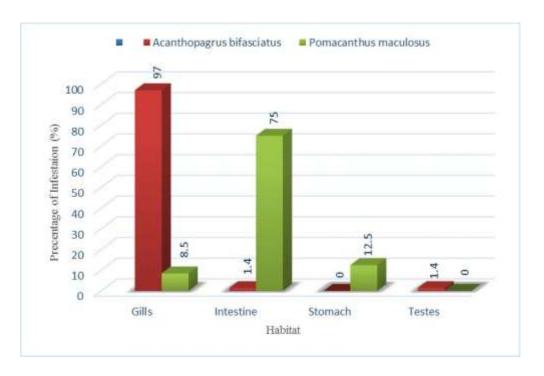


Figure 44. The microhabitat of parasite groups in fish hosts sampled from Qatar.

### **5- Mixed infestation**

Most parasites were found in a single infection, i.e. infection caused by a single parasite species. However, double infestations were detected in both fish hosts:

<u>Acanthopagrus bifasciatus:</u> There was a double infestation on the gills between (Monogenea, and Copepod 12 times) and between (Isopoda and Monogenea three times).

<u>Pomacanthus maculosus:</u> There was a double infestation between Nematoda and Trematoda (one time) in the intestine.

# 6- Host specificity

In the present study, two species of parasite: one monogenean species Polylabris mamaevi and one isopod species Gnathia africana pranazia larva where found to be not specific, as they infested the two-fish hosts

## 7- Relationship between fish size and infection

## 7.1- Relationship between Length and infestation:

Acanthopagrus bifasciatus: By comparing average length and variability of infested versus non-infested specimen of two-bar seabream, no significant difference was found. Figure 45 shows a box-plot of the length of Acanthopagrus bifasciatus among infested and not-infested groups. The results of Shapiro-Wilk test (P> 0.05) for length of infested and not infested groups show that the data is approximately normally distributed and T-Test was used to see if there is a significance difference in Length among infested and non-infested groups. The results of T-test (P> 0.05) show that there is no significance difference in fish length between infested and not infested groups.

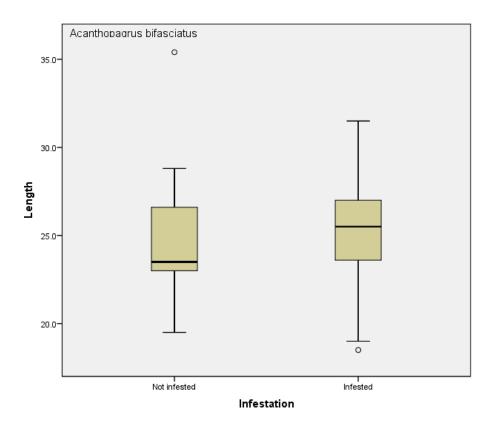


Figure 45. Box-plot showing the length of Acanthopagrus bifasciatus among infested and not-infested groups.

<u>Pomacanthus maculosus:</u> A significant difference in lengths between infested and not infested groups of fishes was found using a T-test (P<0.05). Figure 46 shows a box-plot of the length of *Pomacanthus maculosus* among infested and not-infested groups. The results of Shapiro-Wilk test (P>0.05) for length of infested and not infested groups show that the data is approximately normally distributed and T-Test was used to verify if there is a significance difference in Length among infested and non-infested groups.

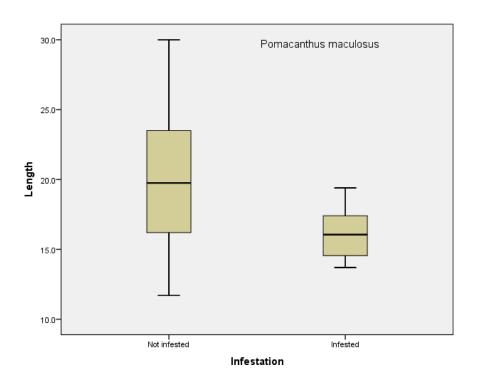


Figure 46. Box-plot showing the length of *Pomacanthus maculosus* infested by parasites among infested and not-infested groups.

# 7.2- Relationship between Length and intensity of infestation:

Acanthopagrus bifasciatus: There is a significant positive correlation between length and intensity of infestation (r = 0.5, P<0.05) (Fig. 47).

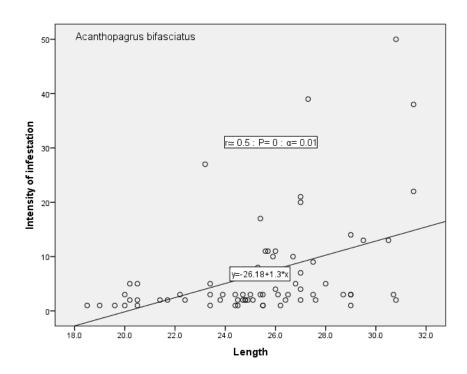


Figure 47. correlation between intensity of infestation (number of parasites in each fish) and length of *Acanthopagrus bifasciatus* sampled from Qatar.

## 7.3- Relationship between weight and infestation:

Acanthopagrus bifasciatus: No significant difference in weight between infested and not infested groups was found using Mann-Whitney U test (P> 0.05). Figure 48 shows a box-plot of the weight of Acanthopagrus bifasciatus among infested and not-infested groups of fishes. The results of Shapiro-Wilk test (P< 0.05) for weight of infested and not infested groups show that the data is not normally distributed and Mann-Whitney U test for non-parametric data was used to see if there is a significance difference in Length among infested and non-infested groups.

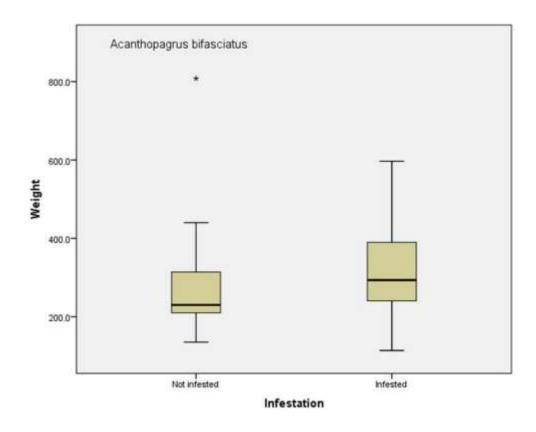


Figure 48. Box-plot showing the weight of Acanthopagrus bifasciatus among infested and not-infested groups.

<u>Pomacanthus maculosus:</u> A significant difference in weight between infested and not infested groups was found using Mann-Whitney U test (P< 0.05). Figure 49 shows a box-plot of the weight of *Pomacanthus maculosus* among infested and notinfested groups of fishes. the results of Shapiro-Wilk test (P< 0.05) for weight of infested and not infested groups show that the data is not normally distributed and Mann-Whitney U test for non-parametric data was used to see if there is a significance difference in Length among infested and non-infested groups.

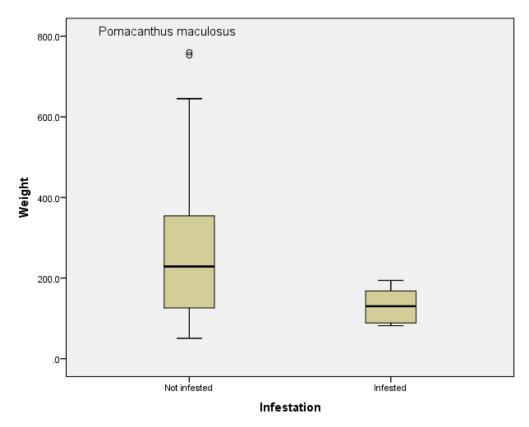


Figure 49. Box-plot showing the weight of *Pomacanthus maculosus* among infested and not-infested groups.

# 7.4- Relationship between weight and intensity of infestation:

<u>Acanthopagrus bifasciatus:</u> there is a significant positive correlation (Fig. 50) between weight and intensity of infestation (r = 0.5, P < 0.05)

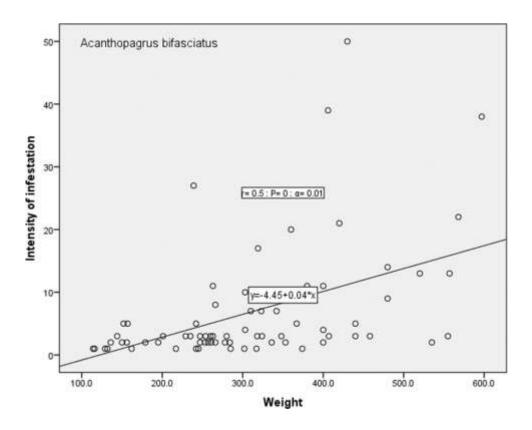


Figure 50. correlation between intensity of infestation (number of parasites in each fish) and weight of *Acanthopagrus bifasciatus* sampled from Qatar.

### 7.5- Relationship between season and infestation (Temporal variation):

<u>Acanthopagrus bifasciatus:</u> The results of Fisher's exact test (P> 0.05) shows that there is no significant difference in infestation between Fall and Spring.

### 7.6- Relationship between season and intensity of infestation:

<u>Acanthopagrus bifasciatus</u>: The intensity of infestation of Acanthopagrus bifasciatus parasites among Spring and Fall is illustrated in a Box-plot (Fig. 51).

The results of Shapiro-Wilk test (P < 0.0) for intensity of infection among Fall and Spring show that the data is not normally distributed.

Mann-Whitney U test for non-parametric data was used to examine if there is a significant difference in intensity of infestation among Spring and Fall. The results of Mann-Whitney U test (P<0.05) show the average intensity of infestation to be significantly larger during Spring than during Fall.

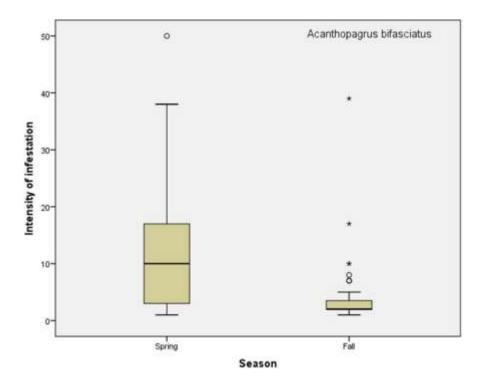


Figure 51. Box-plot showing the intensity of infestation of Acanthopagrus bifasciatus parasites among spring and fall.

## 7.7- Relationship between fish sex and infection:

<u>Acanthopagrus bifasciatus:</u> No significant difference in infestation between male and female was verified using Fisher's exact test (P>0.05).

# 7.8- Relationship between sex and intensity of infestation:

Acanthopagrus bifasciatus: No significant difference in intensity of infestation between mall and female groups was found using Mann-Whitney U test (P>0.05). Figure 52 shows a box-plot that illustrate the intensity of infestation of Acanthopagrus bifasciatus parasites between male and female.

The results of Shapiro-Wilk test (P< 0.0) for intensity of infection between male and female show that the data is not normally distributed.

Mann-Whitney U test for non-parametric data was used to see if there is a significance difference in intensity of infestation between male and female.

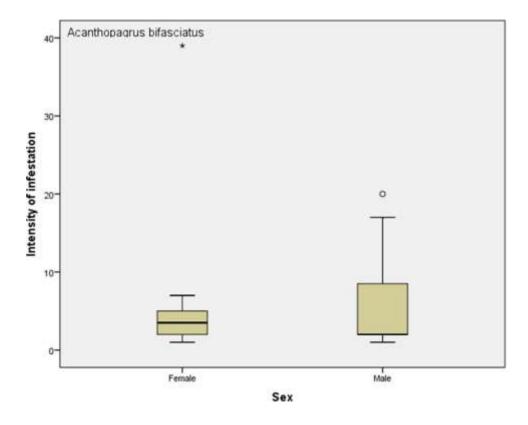


Figure 52. Box-plot showing the intensity of infestation of Acanthopagrus bifasciatus parasites between male and female.

# 8- Spatial variations and fish health

By comparing specimens, the coral reef fish *Pomacanthus maculosus* collected from different locations (Fig. 53) from Qatari water, it was found that the general anatomical condition is almost the same.

Spatial variations in parasitic infection for *Pomacanthus maculosus*:

Out of the five fishes which were examined from Zubara, 3 (60 %) were found to be infected with Nematoda 1 (20 %).

Out of the ten fishes which were examined from Umm Al-Ashran, 2 (20 %) were found to be infected with one or more of the following groups of parasites: Nematoda 2 (20 %) and Trematoda 1 (10 %).

Out of the ten fishes which were examined from North (1) 3 (37.5 %) were found to be infected with Nematoda.

Out of the 11 fishes which were examined from Al-Ashat Island, 2 (18.2%) were found to be infected with one or more of the following groups of parasites: Monogenea 2 (18.2%) and Isopoda 2 (18.2%).

No infection was found in the following locations: Fasht Al-Udayd (5 fishes) and North (3) (one fish).

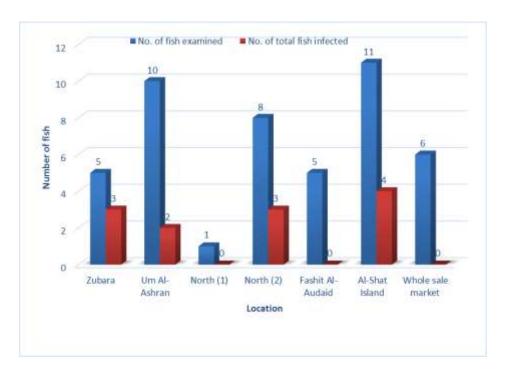


Figure 53. Number of fish of examined and infested fish *Pomacanthus maculosus* sampled from Qatar in each location.

### 9- Gut contents

By examining the gut contents for all the examined fishes, no difference was found in the gut contents between infested and non-infested fish specimens. The gut contents of each species were one or more of the following (some time the gut contents were fully digested and cannot be distinguished:

Acanthopagrus bifasciatus: Small shells, pieces of meat, shrimp larva and pieces of crab.

Pomacanthus maculosus: Small shells, algae, pieces of meat and small star fish.

#### **DISCUSSION**

#### **Anatomical observations**

Fish diseases, including liver pathology can be used as standard indicators for the health of the ecosystem (Stentiford *et al.*, 2009). The aim of the present work was to assess the health status of coral reef associated fishes in Qatari waters. This was done by examining several indicators of fish health, with special emphasis to fish ectoparasites, in Qatari seawaters by examining several indicators related to fish health. Fish health endpoints were assessed by recording abnormalities in representative sampled of fishes using several features, among those: (1) intensive parasitic infestations (fish with two species of parasites or with four parasites or more per one fish) (2) ulcers, (3) tumors, (4) fin erosion and (5) body abnormalities (Moore, *et al.*, 1997). In the present study, no health abnormalities were detected (except intensive parasitic infestations). However, some anatomical observations were noticed, by comparison with the other examined fishes from the same batch (having the same conditions regarding fishing method, handling and degree of freshness).

All the anatomical observations observed on *Acanthopagrus bifasciatus* combined with the parasitological infestation by Monogenea (*Polylabris mamaevi*) during the present study are matching those reported by Bayoumy *et al.* (2015) who made a study on the monogenean parasite *Polylabris lingaoensis* infesting the gills of *Acanthopagrus bifasciatus* from Egypt (Red sea). The authors explained some of the observations as follows: the excessive mucous secretion on gills may be due to the acute irritation, which occurs as a result of parasite infestation (mobility, fixation and feeding) It may also be used by the fish as a mechanical defense, to reduce infestation (Bayoumy *et al.*, 2015). Pale liver and gills may be due to the anemia which takes place as a side-

effect of parasite feeding on fish blood. Moreover, parasitic infestation leads to loss of appetite, which also increases anemia. Injuries in the front of the fish may be due to the attempts of the fish to get rid of the irritation caused by the parasite by scrubbing its body against hard objects.

The abnormal branches in the gills of *Acanthopagrus bifasciatus* which were noticed in the present study, in combination with the copepod parasite *Caligus haemulonis* may be explained by the proliferation of the interlamellar epithelium as a result of parasitic infestation which lead to massive proliferation of mucus cells and lamellar fusion, as suggested by Manera & Dezfuli (2003).

The bile of most teleosts gallbladder contains bilirubin (light yellow) and biliverdin (bluish green) conjugates and less amounts of unconjugated bilirubin. During long time fasts the bile of fish shows markedly increase in the concentration of bilirubin and biliverdin (Cornelius, 1991). During the present study, the colour of the gall bladder bile was light yellow, except in a small number of *Acanthopagrus bifasciatus*, where the colour was dark green. This variation of bile colour may be due to variation in the composition of bile (percentage of bilirubin to biliverdin), which may vary as a result of different feeding history. This observation is in agreement with the findings of Robb (1992), who studied the changes in the gall bladder of the fish (*Merlangius merlangus*) and attributed it to the recent history of feeding. His findings showed that when fish receives regular meals for a period of time, this would lead to a small, pale and empty gall bladder. In contrast, after deprivation of food, the gall bladder increased in weight and the bile gradually changed to dark green then dark blue with time. When the fish fed again, the weight of the gall bladder decreased and the bile started to be discharged after 15 minutes. Those observations were suggested to be applied in field studies to

determine whether the fish stomach contents were lost during capture or empty at capture. (Robb, 1992).

The presence of the unknown balloon-like structure in the gill chamber of *Acanthopagrus bifasciatus* (only one fish out of 89) need further investigations to be explained. Pictures were shared with fish veterinary expert for feedback.

The presence of red dots on the gill of *Pomacanthus maculosus* are likely to be the result of protozoa, bacteria or virus infection but this needs to be verified by further investigations.

The presence of patches on the skin of *Pomacanthus maculosus* without scales was probably an artefact of the sampling method (spearfishing).

The liver colour in most wild fish is light-brown in herbivores and reddish-brown in carnivores and may be yellow in certain times of the year (Taddese *et al.*, 2014). The variation in colour can be related to variation in dietary habits, hepatocyte content, vascularization and health condition (Sales *et al.*, 2017). In the present study, the liver of *Pomacanthus maculosus* showed the presence of different percentages of black sections in the liver ranging from just black coloured edges to totally black liver. This cannot be explained by variation in dietary habits because the fishes (that have different percentages of black sections in the liver) were caught from the same place and had similar gut contents. This observation needs further investigations.

The present study consider the first to record anatomical abnormality of two fish species (*Acanthopagrus bifasciatus* and *Pomacanthus maculosus*) associated to coral reef in Qatari water because the previous studies accomplished about *Acanthopagrus bifasciatus* in Qatari water (El Sayed & Abdel-Bary, 1993 and El Sayed & Abdel-Bary, 1994) were about the biology of the fish and not on their health status. The lack of

background information on anatomical observations of these two species in the region, limited the conclusions this study was able to draw from the observed anomalies.

#### **Intensive parasitic infection**

In the present study, the percentage of fish intensively infested by parasites (fish with two species of parasites or with four parasites or more per one fish) was 37.1% for *Acanthopagrus bifasciatus* and 6.5% for *Pomacanthus maculosus*, but there is no reference number to compare with as all previous studies made on fish parasites of Qatari water (mentioned in detail in the introduction and literature review part) was focusing on the parasite itself not on the fish host.

### Prevalence of the parasites in fish hosts

The total prevalence of parasites in *Acanthopagrus bifasciatus* in the present study was 80.9 %: 76.4% for Monogenea, 12.4% for Copepoda, 3.4 % for Isopoda and 2.2% for Nematoda. There was no infestation by Trematoda or Cestoda. This result is substantially different from the results of previous studies in Qatar, not only in the prevalence but also in the groups of the parasites encountered. The prevalence of Trematoda was reported as 7.7 % (Saoud *et al.*, 1986a) or 8.8 % (Al-Kawari *et al.*, 1996). The prevalence of Cestoda was reported as 8% (Al Kuwari and Kardousha (2002). No infestation with Monogenea, copepoda, isopoda or Nematoda has been previously described. The difference is expected as a result of the long time period between the studies. Regarding previous studies in the Arabian Gulf (Saudi Arabia) the

prevalence was mentioned as 50% for Monogenea, 50% for Crustacea, 40% for Digenea (Bayoumy *et al*, 2012). In other studies, the prevalence was reported as 22.91 % for Monogenea (Hassan *et al.*, 2015) and 38.18 % for Copepoda (Bayoumy *et al.*, 2013) the difference). The variations in prevalence may be due to difference in locality and the parasite species.

The total prevalence of parasites in *Pomacanthus maculosus* in the present study was 26.1 %: 15.2% for Nematoda, 4.4 % for Monogenea, 2.1 % for Trematoda. 4.4 % for Isopoda and no infestations was observed from Cestoda. In the previous studies the prevalence of *Pomacanthus maculosus* parasites was 30% for Cestoda (Al Kuwari and Kardousha (2002) from Qatar and 40% for Nematoda from Iraq (Li *et al.*, 2016) the difference in prevalence may be due to differences in time of study and/or differences in localities of collection.

## Microhabitat of the parasites

In the present study, the common microhabitats of parasites in *Acanthopagrus bifasciatus* were the gills for Monogenea, Copepoda and Isopoda and the intestine and testes for Nematoda. In *Pomacanthus maculosus* the microhabitats were the gills for Monogenea, and Isopoda and the intestine for Nematoda. This is in agreement. The present results are coincide with previous studies (Bayoumy *et al.*, 2013, Hassan *et al.*, 2015 and Li *et al.*, 2016).

#### Mixed infestation

In the present study, the two fish species have single infestation with parasites, but sometimes, *Acanthopagrus bifasciatus* had a double infection in the gills with Monogenea and Copepoda or Monogenea and Isopoda while *Pomacanthus maculosus* had a double infestation in the intestine with Trematoda and Nematoda. This is not in agreement with the previous studies, where all infestations of these two fishes were by a single infestation (Al-Kuwari *et al.*, 2002; Bayoumy *et al.*, 2013; Hassan *et al.*, 2015 and Li *et al.*, 2016).

## **Host specificity**

In the present study, there are two parasites (*Polylabris mamaevi* and *Gnathia africana* pranazia larva) that infested the two fish hosts. These results are backed by previous studies, where those two parasites were not host specific and were found to infest other fish host species (Smit *et al.*, 1999; Tingbao *et al.*, 2007 Adday 2013). Nevertheless, the present study represents the first record of *Polylabris mamaevi* and *Gnathia africana* pranazia larva from *Acanthopagrus bifasciatus* and *Pomacanthus maculosus*, so they are considered new hosts for these parasite species. Moreover, *Acanthopagrus bifasciatus* is considered a new host for another three parasite species *Caligus haemulonis*, *Alella* sp. and *Lernanthropus sarbae*, while *Pomacanthus maculosus* is considered a new host for another parasite *Stephanostomum* sp.

## Relationship between fish size and infestation

The results of the present work showed that in the case of Acanthopagrus bifasciatus, there was no significant differences in size (length or weight) between infested and not infested groups which means that the fish size have no effect on infestation. The opposite is true in the case of Pomacanthus maculosus where there is a significance difference in size (length and weight) between infested and not infested groups, which indicates the effect of fish size on infestation. The obtained results can be explained by the fact that most of the parasites which infested Acanthopagrus bifasciatus were ecto-parasites while most of the parasites which infested Pomacanthus maculosus were endo-parasites. Endo-parasites be largely affected by the immune system of the fish, which will help healthy fish to get rid of parasites and not to develop infestation and the health status of the fish is indicated by its size so it will be expected that large sized fish is not get infested easily. The opposite is true for ecto-parasites. Instead, they will be affected by the environmental conditions surrounding the fish and the parasites, so the size of the fish will not have noticeable effect on infestation.

In the case of *Acanthopagrus bifasciatus* there is a significant positive correlation between the size (length and weight) and intensity of infestation. (the number of infested fish in the case of *Pomacanthus maculosus* was too small to perform the correlation analysis). The above result is expected because, once the fish get infected, larger fish will tend to have a greater number of parasites because it offers larger space and more food for parasites, which make it more suitable as a habitat for developing parasites. This explanation is in agreement with the findings of Barber & Poulin (2002), who confirmed that the importance of the fish host body size come from its effect on diet and determining the size of the available space for colonization. The

results are also agreed with de la Cruz *et al.* (2013) who found that larger fish (Nile tilapia) were infested by greater number of parasites (*Acanthogyrus* sp.) than smaller fish (i.e., there was a positive correlation between size of the fish and intensity of infestation). Ibrahim (2012) explained the positive correlation between intensity of infestation and fish length by the concept that larger fish is older in age and had longer time to harbour parasites than smaller (younger) ones.

### **Relationship between season and infestation (Temporal variation)**

The results of the present work showed that in the case of *Acanthopagrus bifasciatus* there is no significant difference in infestation between Fall and Spring collections but there is a significant difference in intensity of infestation between fall and spring, with larger intensity of infestation occurring more in Spring than in Fall. However, for *Pomacanthus maculosus*, there was no samples during Fall to compare. Ramadan (1991) found that the highest infestation value was detected in winter followed by spring then fall and finally summer but she did not make any statistical analysis to test the significance of her data.

## Relationship between sex and infestation

The results of the present work showed that in the case of *Acanthopagrus* bifasciatus there is no significant difference in infestation and intensity of infestation between male and female (the number of *Pomacanthus maculosus* that could be sexed was too small to do the test). In contrast, Ibrahim (2012) found a positive correlation between fish sex and parasite intensity in *Tilapia zilli* and suggested that the presence of more parasites in females than in males takes place during the period of gonads

development, where females invest more energy in reproduction than males, which will make them in consequence more susceptible to parasite infestation.

#### **Spatial variations and fish health**

The general anatomical condition of the coral reef fish *Pomacanthus maculosus* specimens collected from different locations from Qatari water was almost the same but the species of parasites infested the fish varied from one locality to another. This is expected, due the variation in environmental conditions in each location.

#### **Gut contents**

No difference was found in the contents of the gut between infested and non-infested fish specimens. This may indicate that parasites were not transmitted through a unique kind of food. In the present study gut contents in *Acanthopagrus bifasciatus* was mainly formed by: small shells, pieces of meat, shrimp larva and pieces of crab. The gut contents of *Pomacanthus maculosus* were composed of small shells, algae, pieces of meat and small star fish. This result is in agreement with Torquato *et al.* (2017), who described these two species as invertivore and omnivore, respectively.

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