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First record of two parasitic species *Microcotyle erythrini* and *Clavellotis briani* on the gills of *Dentex macrophthalmus* Fish in the Coastal Water of Lattakia, Syria

Taghrid Layka¹, Badr Al-Ali², Muhammad Nassif³

ABSTRACT

The current study aimed to investigate the infection with Monogenea ectoparasites that infect Dentex macrophthalmus in the Syrian Marine Water and to determine the prevalence rates and mean intensity of infection isolated from this fish, which in turn contributes to the enrichment of the database of fish parasites in the Syrian Marine Water. Where, 115 individuals were collected from the locations of Ras Al-Baseet and the Port of Fishing during the period from the beginning of June 2021 to the end of June 2022. The external organs (gills, oral cavity) of all caught fish samples were examined to detect infection with external parasites. In this research, the two parasitic species Microcotyle erythrini (Microcotylidae) and Clavellotis briani (Lernaeopodidae) were isolated from the gills of Dentex macrophthalmus fish, where the number of infected fish with these two parasitic species was 42 out of 115 Dentex macrophthalmus fish examined. The number of fish infected with the parasitic species M. erythrini was 36 fish distributed as follows: (15, 21) infected fish at the locations of Ras Al-Baseet, the Port of Fishing respectively, with an average total rate of infection (31.16%) at the location of Ras Al-Baseet and (41.65%) at the location of the port of Fishing, while the total intensity of infection was similar at both locations with an average of (1.23, 1.33) parasite/fish at Ras Al-Baseet, the Port of Fishing respectively. The number of infected fish with the parasitic species C. briani was 6 fish distributed as follows: (1, 5) infected fish in the locations of Ras Al-Baseet, the Port of Fishing respectively, with a total rate of infection (12.5%) and (53.3%) and a total intensity of infection (1, 2.6) parasite/fish at Ras Al-Baseet, the Port of Fishing respectively. This is the first time that these ectoparasites have been isolated from Dentex macrophthalmus fish in the Syrian Marine Water. The highest Percentage and Intensity of infection were recorded during summer and spring for: Microcotyle erythrini, Clavellotis briani in the both locations.

Keywords: Syrian Marine Water, *Dentex macrophthalmus*, *Microcotyle erythrini*, *Clavellotis briani*.

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1. INTRODUCTION

Fish is considered an important source of many vital amino acids and vitamins such as: (A, D, E) and some minerals such as: Iodine, Phosphorus and Magnesium. In addition of containing an amount of necessary fats, especially Omega-3 which contributes in developing and growth of the human body (Hibbeln and Davis, 2009). Fish play a role of holding and conducting of Pathogens which are transmitted by Aquatic Environment. Out of these Pathogenes: Viruses, Germs, Fungi and Parasites. Some of these parasites and their stages can move to human beings through eating raw or undercooked fish (Ruppert and Barnes, 1994; Castro et al., 2004).

Parasitic diseases that are considered infectious diseases account for about 80% of the warm water diseases caused by protozoa or Monogenea parasitic (Duijn, 1956; Fijan, 1983). External or Internal Parasites play an important role in the life of fish, as they effect on the immunity of infected fish and make them more susceptible to other diseases such as: Parasitic and bacterial diseases, etc. (Hoffmann, 1999). Sparidae Family is considered one of the high economic and nutritional value fish and it forms (21.5) % of Artisanal Fishing in the Syrian Marine Water (Ulman et al., 2015). *D. macrophthalmus* is considered one of the fish that is economically important (Saad, 2005). It spreads widely all over the Mediterranean. This species feeds on the Crustaceans and Mollusks (Anderson, 2000).

In Syria, many studies were conducted especially on the Fresh Water Fish, but studies on the Marine Fish Parasites were few compared to the number of the Marine Fish found in the Syrian Marine Water. In a study conducted on two kinds of fish: *Siganus rivulatus* and *Siganus luridus*, out of them two parasites were isolated: *Glyphidohaptor plectoccir*, *Gnathia sp* (Hassan et al., 2010). The researcher Hassan et al., (2017) isolated a type of external parasite (*Furnistinia echeneis*) which is of Diplectanidae Family. This parasite was isolated from two types of fish (*Sparus aurata*, *Boops boops*) in the Syrian Marine Water; the highest rate of infection was recorded in the spring and the lowest in the winter.

Hassan et al., (2021) isolated *Hysterothylacium aduncum* from the inferior part of intestine of two fish species *Saurida undosquamis* and *Stephanolepis diaspros* in the Syrian Marine Water. Layka et al., (2016) were able to record the presence of the parasite (*Grubea cochlear*) in (*Mullus surmuletus*) Fish which belongs to the Mazocraeidae Family for the first time in the Syrian Marine Water in the Mediterranean. The researchers Layka and Badran, (2018) were able to record the presence of the Parasitic (*Microcotyle mugilis*) which belongs to Monogenea Class in (*Siganus rivulatus*) Fish cultured in Al- Sin farm, Banias.

These two researchers were also able to isolate the parasite (*Ligophorus Cephalic*) for the first time from the gills of *Liza aurata* Fish cultured at Al-Sin Marine Farm (Layka and Badran, 2019). In a study conducted in the Syrian Marine Water, the parasitic (*Lamellodiscus elegans*) was isolated from the gills of (*Lithognathus mormyrus*) (Hassan et al., 2018).

Research objectives and importance

As a result of the increasing demand for marine fish as a food source rich in animal protein and unsaturated fatty acids on the one hand and the lack of marine fish resources on the other hand. The importance of the research is clear in the study of ectoparasites of *Dentex macrophthalmus* fish, which is one of the species belonging to the Marine Sparidae Family and is one of the economically important fish species as well as its importance in the field of aquaculture in the countries of the Mediterranean Basin, as it is easy to breed and handle.

This makes it possible to know the effect of these parasites on this type of fish. All of this evoked for the study of all aspects related to this type of fish found abundantly in the Syrian Marine Water and the study of the relationship between parasites and the bio morphometric characteristics of the fish of this type in the marine water of Lattakia at the two locations of the Port of Fishing and Ras Al-Baseet and thus contributing to the development of ways to reduce the infection of fish with these pathogens and its prevalence.

The study aims to:

Investigation of infection with ectoparasites of *Dentex macrophthalmus* fish of Sparidae family in Lattakia city beach. Studying the temporal (seasonal) and spatial changes in the rates and intensity of infection with these parasites.

2. METHODS AND MATERIALS

115 individuals of *D. macrophthalmus* (Figure 1) were collected from two locations: Ras Al-Baseet and the Port of Fishing (Figure 2, 3), during the period from the beginning of June 2021 to the end of June 2022 and were transferred to the laboratories of the Higher Institute for Marine Research in Tishreen University for the purpose of laboratory examination and detection of parasites.

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Figure 1 Large-eye dentex sample

The location of Ras Al-Baseet

It is located 40 km north of Latakia within the following coordinates: 35°50′ 52.7485″ E, 35°51′9.9634″ N. It is a wide bay with black sands, preceded by an elongated tongue to the west with zigzag sides. The area is characterized by a relatively low population density and human activities are limited to fishing and other tourism activities, which makes this location relatively less vulnerable to pollution compared to other areas. It is also characterized by the presence of a single port of fishing.



Figure 2 The location of Rass Al - Baseet

The location of the Port of Fishing

It is located in Lattakia within the following coordinates: 35°46′ 20″ E, 35°32′15″ N. The area of this location is approximately 2 km and the depth of the water column ranges between (3.5-16) m. In this location, there are three outfalls of sewage, spread over a length of 1 km. One of the most important features of this location is that it is the entrance to the port of Lattakia, which is completely restricted from all sides (Figure 3). In addition to being affected by household waste transported through the sewage water of the city of Lattakia, it is therefore considered a polluted location.

After the samples were transferred to the laboratory, the fish were washed with sterile physiological saline to remove sand and dirt and then some morphometric measurements (length, weight, age) were taken. The age of the fish was determined using scales by removing (5-6) scales from the area above the lateral line and below the dorsal fin and then washed with water and placed in a 4% NH4OH ammonium aqueous solution for two minutes, then washed and kept between two glass slides, in order to identify the growth rings using a magnifying glass according to (Sarder, 2015).

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Figure 3 The location of the Port of Fishing

Swabs were taken from different parts of the fish's body (skin, gills and oral cavity) and examined carefully under the naked eye and under a magnifying glass first to detect pathological features. While the parasites were removed from different parts of the body of the fish using forceps, placed on clean slides and then examined under a microscope with different magnifications and pictures of the parasitic preparations were taken. Parasites were fixed using alcohol 70%. Isolated parasitic species were identified based on the reference taxonomic keys (Pritchard and Kruse, 1982; Lucky, 1977; Amlacher, 1976). The rate and intensity of infection were also determined based on the following two relationships (Margolis et al., 1982; Bush et al., 1997).

Prevalence % = (number of infected fish /number of fish tested) * 100.

Intensity = number of isolated parasites /number of infected fish.



Figure 4 Weighing of a Fish Sample



Figure 5 Length of a Fish sample

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3. RESULTS AND DISCUSSION

In the current study, the two parasite species *Microcotyle erythrini* which belongs to the family Microcotylidae and the species *Clavellotis briani* which belongs to the family Lernaeopodidae were isolated and identified from the gills of *D. macrophthalmus* Fish for the first time in the Coastal Water of Lattakia City.

Table 1 Classification of the first type of Monogenea parasites by Worms taxon (https://www.marinespecies.org/)

	Microcotyle
	erythrini
Kingdom	Animalia
Phylum	Platyhelminthes
Class	Monogenea
Order	Mazocraeidea
Family	Microcotylidae
Genus	Microcotyle
Species	M. erythrini

1-Microcotyle erythrini

General Description

The mouth is sub-ventral, conical in shape and provided with a pair of separate hollow suckers. The pharynx is semi-spherical, the esophagus is short and the intestine bifurcates from the posterior side to give what is known as the genital atrium. Caeca extend to the stabilization member and sometimes to the posterior peduncle with many complex internal and external lateral branches. Multiple testicles, numbering from (12-20) testicles, flattened from the dorsal-abdominal side, semi-elliptical to irregular, most of them anteriorly positioned, arranged in bunches (1-2) row (Villora-Montero et al., 2020).

The vas deferens is relatively straight; the organ of sexual intercourse is muscular and is located in the posterior part of the genital atrium. The genital atrium is composed of a muscular chamber that is transversely located in the middle and is equipped with small conical spines, numbering between (216-408) forks. The oviduct is slightly tortuous and includes an elongated seminal vesicle that extends towards the anterior-arterial region. To end with the Ootype ovary, the vitelline glands are well developed (Víllora-Montero et al., 2020).

The vitelline ducts are Y-shaped with two separate efferent ducts (left and right) joined together laterally abdominal at the level of the germarium. The eggs are spindle-shaped with two threads. The outer thread in the ovipositor is long and thin, thickening at the posterior end, while the inner thread is shorter and has a hard, pointed end (Víllora-Montero et al., 2020).

Prevalence and Intensity of infection

The number of infected fish with the type *Microcotyle erythrini* was 36 *Dentex macrophthalmus* Fish, distributed as follows: (15, 21) infected fish at the locations of Ras Al-Baseet and the Port of Fishing, respectively.

The average percentage of total infection of *Dentex macrophthalmus* Fish with the parasitic species *Microcotyle erythrini* was (31.16%) at the location of Ras Al-Baseet and (41.65%) at the location of the Port of Fishing, while the average intensity of infection was similar at both locations (1.23, 1.33) parasite/fish at Ras Al-Baseet, the Port of Fishing respectively.

At the location of Ras Al-Baseet, the highest rate of infection was in the summer (57.14) % and the lowest rate of infection was recorded in the winter (11.1%). The highest intensity of infection was recorded in the summer (1.5) parasite/fish and the lowest intensity of infection was recorded in the winter (1) parasite/fish.

The highest rate of infection at the location of the port of fishing was in the summer (66.6%) and the lowest was in the winter (20%), while the highest intensity of infection with this parasite in the summer was (1.83) parasite/fish and the lowest in the winter was (1) parasite / fish and this indicates the predilection of this parasitic species and the association of infection with it with warmer water temperatures.

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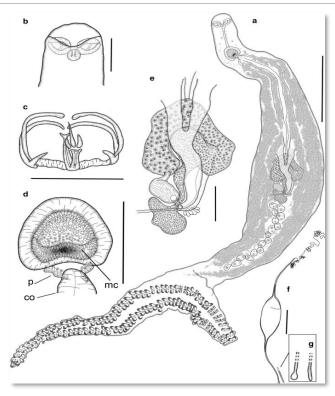


Figure 6 Diagram represents Microcotyle erythrini

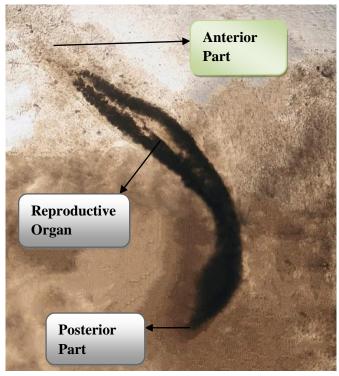


Figure 7 General Form of Microcotyle erythrini

This was confirmed by Gasmi et al., (2017) in their study conducted in Al Qalaa Coast, Algeria, in which the parasitoid genus *Choricotyle sp.* which the presence of the parasite was more noticeable in the warmer seasons, as the rate of infection was highest in the summer and then in the spring to a lesser extent. *Microcotyle erythrini* was also isolated in a study by Bouguerche et al., (2019) from the gills of *Pagellus erythrinus*. In 2018 study on *Lithognathus mormyrus*, in which the parasitic species *Microcotyle spinicirrus* was isolated during the spring, as the number of isolated parasites reached 6 parasites from the first and fourth gill arches of individuals, with the rate of infection (15.38%).

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Table 2 The number of *Dentex macrophthalmus* fish infected with *Microcotyle erythrini* and the number of parasites isolated from them at the location of Ras Al-Baseet

Intensity	Dwarralan aa 9/	The number of	The number of	The number of	Coocon
(Parasite/ fish)	Prevalence %	parasites isolated	infected fish	fish studied	Season
1	11.1	1	1	9	Winter
1.3	37.5	4	3	8	Spring
1.5	57.14	6	4	7	Summer
1.14	18.91	8	7	37	Autumn
4.94	124.65	19	15	61	Total
1.23	31.16				Average

Table 3 The number of *Dentex macrophthalmus* fish infected with *Microcotyle erythrini* and the number of parasites isolated from them at the location of the Port of Fishing

Intensity	Prevalence %	The number of	The number of	The number of	Season
(Parasite/ fish)	Prevalence %	parasites isolated	infected fish	fish studied	
1	20	1	1	5	Winter
1.4	50	7	5	10	Spring
1.83	66.6	11	6	9	Summer
1.1	30	10	9	30	Autumn
5.33	166.6	29	21	54	Total
1.33	41.65				Average

The current study agreed with the results of the study conducted in Turkey on some fish belonging to the family Sparidae: *Dentex dentex, P. erythrinus, Boops boops, L. mormyrus,* where the parasitic species *M. erythrini* was isolated from the gills of these fish and the increase in rates of the percentage and intensity of infection with this type of parasite on the studied fish during the summer and spring seasons and its gradual decrease in the autumn and winter seasons.

Our results also agreed with a study conducted by Ramadane et al., (2013) in Algeria, in which the parasitic species *Microcotyle erythrini* was isolated from the gills of two species of fish, *Boops boops*, *Mullus barbatus*, where the highest rate of infection with this parasite was recorded in the fish *Boops boops* in the spring, to gradually decrease in the autumn and winter seasons, while the highest percentage of infection with this parasite was recorded in *M. barabatus* fish in the summer (August), when it reached (27%).

Table 4 Classification of the second type of Monogenea parasites by Worms taxon (https://www.marinespecies.org/)

	<u>Clavellotis briani</u>
Kingdom	Animalia
Phylum	Arthropoda
Class	Monogenea
Order	Siphonostomatoida
Family	Lernaeopodidae
Genus	Clavellotis
Species	Clavellotis briani

2-Clavellotis briani General Description Female

The body consists of the head, thorax, cylindrical in shape, a dorsal shield appears from the front and at the level of the upper jaw there are two prominent bubble-like bulges contracting under the head. The posterior angles (edges) of the trunk have a consistently sharp projection which is a taxonomic feature of the species. The genitals are elongated and large. The antennae in the second joint bear soft pointed hairs, while short hairs are observed in the third joint from the ventral side. The head part consists of six hairs of different shapes.

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The antennae consist of an outer region known as an exopodite with an extended circular end without any embellishments. The inner part of the antennae, known as the endopodite, is articulated, bearing a single capillary with a white encircled base. The lower jaw contains five main teeth, in addition to three base teeth and three accessory teeth located between the first four teeth. Endopodite is semi-rectangular, bearing two cylindrical papillae topped by tapering hairs. There are also some vertebrae at the base of the endopodite from the dorsal side.

The exopodite from the lateral ventral side is conical in shape and bears two hairs, one strong cephalic and the other shorter and thinner subcephalic. The upper jaw has a strong structure that bears a group on its inner edge and the articulated part on the body carries a small bristle on its ventral side that ends with a strong apical hook. The female of *Clavellotis briani* measures 4 mm to the end of the genital tubercle (Benmansour et al., 2001).

The male

The antenna has six head hairs of different shapes and sizes. The antennae of the male, unlike the female, have an exopodite that contains a number of vertebrae. Endopodite has three strong hairs at its apex as well as several small spines arranged in a horizontal direction. The upper jaw in the male has a similar structure to the female, but differs from it in terms of the ventral position of the exopodite, in addition to the absence of vertebrae at the base of the endopodite. The male of *Clavellotis briani* measures 0.8 mm. (Benmansour et al., 2001).

Prevalence and Intensity of infection

The number of samples infected with the *Clavellotis briani* species was 6 of *Dentex macrophthalmus* Fish, distributed as follows: (1, 5) infected fish at the locations of Ras Al-Baseet, the Port of Fishing, respectively, with a total rate of infection of (12.5, 53.3) % at the two previous locations, respectively, with the intensity of total infection (1, 2.6) parasite/fish, respectively.

The parasitic species *Clavellotis briani* was isolated at location of Ras Al-Baseet during the spring season only, where the rate of infection was (12.5%) and the intensity of infection was (1) parasite/fish. As for the location of the Port of Fishing, it was isolated during the spring and summer seasons, where the percentage of infection with this parasite in the spring was (20%) with an intensity of infection of (1) parasite/fish and in the summer the rate of infection was (33.3%), with an intensity of infection of (1.6) Parasite/fish.

Comparing the results of our study with the study of Gasmi et al., (2017), in which the parasite *Clavellotis sp.* was isolated from the gills of *Pagellus erythrinus* in El-Qalaa coast, Algeria, members of this parasitoid genus were more frequently seen during the warmer seasons and recorded the highest rates of infection during the spring and summer seasons.

This was confirmed by Boualleg et al., (2018) in their study conducted in Algeria in which the parasite *Clavellotis sp., Clavellotis sargi, Clavellotis pagri, Clavellotis strumosa* were isolated from the gills of *Diplodus annularis, Pagellus erythrinus, Lithognathus mormyrus, Mullus barbatus*. The highest values for the rates of infection were recorded in the summer and spring seasons, as they were (33, 25%) in each of the summer and spring seasons, respectively.

Clavellotis strumosa was isolated in the study by Oktener et al., (2008) in the Sea of Marmara in Turkey from the gills of Pagellus erythrinus Fish during the spring and summer seasons (from May to August), with the rate of infection (6%) and an intensity of infection (1.6) parasite/fish. The study of Koyuncu et al., (2015) in the northeastern Mediterranean in Turkey showed that the parasite Clavellotis briani was isolated from the gills of Lithognathus mormyrus in spring, which in turn agrees with the results of the current study.

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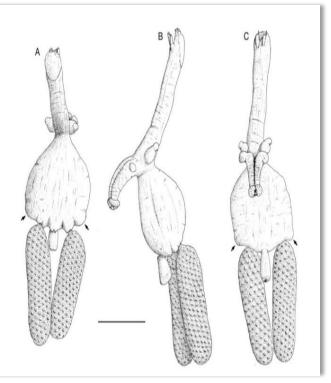


Figure 8 Diagram represents different positions of Clavellotis briani

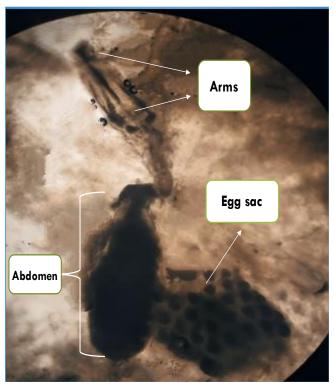


Figure 9 General Form of Clavellotis briani Female

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Table 5 The number of *Dentex macrophthalmus* fish infected with *Clavellotis briani* and the number of parasites isolated from them at the location of Ras Al-Baseet

Intensity	Prevalence %	The number of	The number of	The number of	Season
(Parasite/ fish)		parasites isolated	infected fish	fish studied	Season
0	0	0	0	9	Winter
1	12.5	1	1	8	Spring
0	0	0	0	7	Summer
0	0	0	0	37	Autumn
1	12.5	1	1	61	Total

Table 6 The number of *Dentex macrophthalmus* fish infected with *Clavellotis briani* and the number of parasites isolated from them at the location of the Port of Fishing

Intensity	Prevalence %	The number of	The number of	The number of	Concor
(Parasite/ fish)		parasites isolated	infected fish	fish studied	Season
0	0	0	0	5	Winter
1	20	2	2	10	Spring
1.6	33.3	5	3	9	Summer
0	0	0	0	30	Autumn
2.6	53.3	7	5	54	Total

4. CONCLUSIONS AND RECOMMENDATIONS

- 1. Continue the study to determine the Endoparasitic species of the studied Fish species, in addition to continuing the study of the Marine Fish Parasites to establish a taxonomic database for the Marine Fish Parasites.
- 2. Studying the effect of parasites on the productivity of these economically important fish so that they form a scientific base that can be used later when cultivating these fish.
- 3. Follow-up research related to this type of fish over the years to find out the latest developments.

Informed consent

Not applicable.

Ethical approval

The Animal ethical guidelines are followed in the study for species observation & identification.

Conflicts of interests

The authors declare that there are no conflicts of interests.

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Data and materials availability

All data associated with this study are present in the paper.

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