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Detection and Morphological Assessment of Parasitic Helminths in *Sarpa salpa* and *Euthynnus alletteratus* from the Misurata Local Market

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ABSTRACT

Background and Objective: Parasitic helminths pose a significant threat to marine fish populations by increasing mortality, reducing productivity, and impairing growth; however, information on how feeding habits influence infection patterns remains limited in the Misurata Region. This study aimed to assess the prevalence, distribution, and pathological impact of helminth parasites in two marine fish species with contrasting diets: the carnivorous *Euthynnus alletteratus* and the herbivorous *Sarpa salpa*. **Materials and Methods:** A total of 140 fish specimens (70 per species) were randomly collected from the Misurata local fish market. The digestive tract and gills were examined for helminth parasites using standard parasitological and histopathological techniques. Parasites were identified morphologically, and infection prevalence, site specificity, and tissue damage were recorded. Differences in infection rates between species were statistically evaluated to determine the influence of feeding habits on parasitic burden. Data analysis was performed using SPSS version 26, applying independent two-tailed t-tests at $\alpha = 0.05$. **Results:** Overall, 71.4% of the examined fish were infected with helminth parasites. *Euthynnus alletteratus* exhibited a significantly higher infection rate (70%) compared to *Sarpa salpa* (30%). Combined infections of the gastrointestinal tract and gills were more frequent in *E. alletteratus*, whereas single-site infections predominated in *S. salpa*. Severe histopathological alterations, particularly in the digestive tract, were more pronounced in *E. alletteratus*. The study documented a diverse helminth fauna, including the first regional record of Acanthocephala and a digenean species in *E. alletteratus*, as well as nematode larvae in *S. salpa*. **Conclusion:** The findings demonstrate that helminth infection prevalence, distribution, and severity are strongly influenced by host feeding behavior. This study provides the first baseline data on helminth parasitism in these marine fish species from the Misurata area. Future research should expand seasonal sampling and incorporate molecular identification to better understand parasite transmission dynamics and ecological impacts.

KEYWORDS

Endoparasites, *Sarpa salpa*, digenea, prevalence, identification, *Euthynnus alletteratus,* monogenea

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INTRODUCTION

Parasitic helminths are among the most economically significant parasites affecting fish globally. They infect freshwater, marine, and brackish-water fish species, often causing severe harm to their hosts. Marine fish, in particular, serve as common hosts for diverse digenetic parasites found worldwide. These parasites use fish as intermediate or temporary hosts, infesting all organs, with predatory fish typically harboring heavier infections. Their pathogenic effects vary; some invade the gastrointestinal tract of marine fish, stunting growth and triggering diseases that can be fatal, while others have minimal impact[1,2].

Research from various global regions indicates that parasitic groups such as Monogeneans, Digenean Trematodes, Nematodes, and Cestodes frequently infect marine fishes. Monogeneans primarily infest the gills, skin, and fins of fish. Digenean trematodes exhibit a complex life cycle involving multiple hosts, with fish acting as either primary or intermediate hosts depending on the species. These trematodes can inhabit external surfaces or internal organs, though most species have limited pathogenic effects on their hosts. Nematodes, on the other hand, can invade any organ, often impairing the function of infected tissues. Cestodes typically target the digestive tract, muscles, or other internal organs, with their larval forms (Plerocercoids) ranking among the most harmful fish parasites globally[3].

The Mediterranean Sea has been a focal point for studies on helminth parasites in sparid fish, with extensive contributions from numerous parasitologists over several decades[4,5]. While Gargouri Ben Abdallah *et al*.[6] have documented various helminth species in the region, research remains notably scarce along Libyan coastal waters, with Elmajdoub *et al*.[7] representing one of the few exceptions.

Despite environmental changes that have diminished parasitic infections, Mediterranean marine ecosystems continue to support diverse parasite communities and remain susceptible to various parasitic diseases. This persistence is particularly concerning given the absence of comprehensive studies on helminth infections in Libyan coastal fish species-a significant knowledge gap considering the ecological and economic importance of these marine resources. Understanding helminth diversity and infection patterns is essential for advancing fish health management and sustainable aquaculture practices.

Among the region's ecologically important species, *Sarpa salpa* (Sparidae) stands out for its diversity along the Libyan Coast[8]. Known locally as "Shalba" in Libya and Tunisia[9], this species shares its habitat with *Euthynnus alletteratus* (Scombridae), an opportunistic carnivore commonly called little tunny or bluefin tuna[10,11].

Parasitic infections in marine fish, including *Sarpa salpa* and *Euthynnus alletteratus*, have been extensively studied worldwide. For instance, Antar and Gargouri investigated these infections in the Bay of Bizerte, Tunisia[12], while Mele *et al*.[13] documented parasitic infestations in *E. alletteratus* from the southeastern Iberian coast (western Mediterranean), reporting infections by two monogeneans, four didymozoid trematodes, and four copepods. Similarly, Abdel-Gaber *et al*.[14] identified four trematode species (order Plagiorchiida, three families) in *E. alletteratus* sampled off the Abu Qir landing site, Egypt. Variations in infection rates among these studies likely reflect ecological differences across sampling sites, as environmental conditions strongly influence parasite prevalence[15].

Against this backdrop, the present study aimed to assess the prevalence of helminth infections and morphologically characterize helminth species infecting *Sarpa salpa* and *Euthynnus alletteratus* obtained from local fish markets in Misurata, Libya.

MATERIALS AND METHODS

Study area: Misurata is a coastal city located in Northwestern Libya along the Mediterranean Sea, approximately 200 km East of Tripoli and 257 km West of Sirte. The region experiences a typical Mediterranean climate, characterized by hot, dry summers and mild, wet winters, with an annual mean temperature of approximately 20°C.

Sample collection: A total of 140 fish specimens were randomly collected from March to September 2023 at the Qasr Ahmed local fish market in Misurata, Libya. The samples consisted of 70 individuals from each species, *Euthynnus alletteratus* and *Sarpa salpa*. Immediately after collection, the specimens were placed in insulated containers with ice and transported to the research unit at the Faculty of Science, Misurata University, for further processing.

Species identification and biometric assessment: Initial identification of fish species was conducted based on standard morphological characteristics. Taxonomic confirmation was provided by Dr. Sara Ali Elmabrouk, a Marine Biology specialist at Omar El-Mukhtar University. Each specimen underwent biometric assessment, including measurement of total length (to the nearest 0.1 cm), recording of body weight (to the nearest 0.1 g), and external examination for visible parasitic infections[16].

Parasitological examination: Fish were dissected following standard parasitological procedures. The gastrointestinal tract and gill arches were systematically examined for helminth parasites. Infected tissues were isolated and preserved in 90% ethanol, and organs were separated into sterile Petri dishes. Gentle scraping was performed using distilled water for fragile specimens and 0.9% saline solution to preserve parasite structure. Microscopic examination was conducted at magnifications ranging from 40× to 400×.

Parasite processing and identification: Recovered parasites were collected using sterile plastic pipettes and fixed in 70% ethanol in pre-labeled glass vials. Specimens were stained with Acetic Acid Alum Carmine[17], dehydrated through a graded ethanol series (70, 80, 90, and 100%), cleared in xylol, and mounted on glass slides using DPX mounting medium for morphological identification.

Statistical analysis: Data analysis was performed using SPSS version 26 (IBM Corp.). Independent two-tailed t-tests were applied at a significance level of $\alpha = 0.05$ to compare infection prevalence between species, differences in infection intensity, and relationships between host size and parasite load.

Ethical approval statement: This study was conducted following the Declaration of Helsinki and received formal ethical approval from the Research Ethics Committee at the Faculty of Science, Misurata University. Data collection adhered to the guidelines established by the committee.

RESULTS AND DISCUSSION

This study analyzed 140 bonefish purchased from a local market in the Qasr Ahmed area. The sample included two species: *Euthynnus alletteratus* (weighing between 840 and 1320 grams) and *Sarpa salpa* (weighing between 235 and 350 g). The researchers discovered that while a fish's weight did not predict its likelihood of being infected by parasites, heavier fish tended to have more severe infections. Overall, 71.4% of the sampled fish were infected with parasitic helminths in their digestive tracts or gills. Infection was more common in *Euthynnus alletteratus* (70%) than in *Sarpa salpa* (30%). This infection rate is lower than that reported in a previous study conducted in the Tokra area; this discrepancy may be due to the smaller sample size in the current study. Infections were significantly more prevalent in the digestive tract, with 100% of the infected fish showing signs of infection in this area, compared to 70% in the gills. Statistical analysis confirmed a significant difference in infection rates for *Euthynnus alletteratus* ($p = 0.002$), while no significant difference was found for *Sarpa salpa* ($p > 0.05$) (Table 1).

Table 1: Prevalence rates of helminth infections in *Sarpa salpa* and *Euthynnus alletteratus*

	Euthynnus alletteratus			*Sarpa salpa*			
Study fish type	Gill	Gut	Total	Gill	Gut	Total	Total
Number of infected fish	10	30	30	60	70	70	100
Prevalence	10%	30%	30%	60%	70%	70%	71.4%

Table 2: Prevalence rates of common and independent infections in *Sarpa salpa* and *Euthynnus alletteratus*

	Euthynnus alletteratus		*Sarpa salpa*	
Study fish type	Joint infection	Independent infection	Joint infection	Independent infection
Number of infected fish	1	2	6	1
Prevalence	33.3%	66.7%	90%	10%

Table 3: Intensity of helminth infections in *Sarpa salpa* and *Euthynnus alletteratus*

	Euthynnus alletteratus			*Sarpa salpa*			
Study fish type	Gill	Gut	Total	Gill	Gut	Total	Severity of the total
Intensity No.	15	35	50	40	1218	1258	1308
Prevalence	%30	%70	%3.82	%3.17	%96.8	%96.2	

Analysis of infection patterns showed that co-infection of both the digestive tract and gills was the most common occurrence, found in 70% of parasitized fish. The remaining 30% exhibited an independent infection confined solely to the digestive tract. There was a notable variation between species. *Euthynnus alletteratus* had a significantly higher rate of combined infections, at 90%, compared to *Sarpa salpa*, which had a rate of only 33.3%. Conversely, *Sarpa salpa* displayed a higher prevalence of independent infections (66.7%) compared to *E. alletteratus*, which had only 10%. Remarkably, all but one *E. alletteratus* had a combined infection, while all but one *S. salpa* had an infection confined to the digestive tract. Despite these descriptive trends, statistical analysis indicated that the differences between the rates of independent and combined infections within each species were not significant ($p>0.05$). The tendency for carnivorous fish like *E. alletteratus* to host combined infections is likely influenced by their dietary exposure to parasites (Table 2).

The total number of parasites, indicated by infection intensity, varied significantly between the two species studied. Researchers isolated a total of 1,308 helminths. The vast majority, 1,258, were found in *Euthynnus alletteratus*, while only 50 were found in *Sarpa salpa*. Additionally, most of the parasites (1,253) were located in the digestive tract, compared to just 55 in the gills. Despite this significant numerical difference, statistical analysis revealed that the variation in infection intensity between the two species was not statistically significant ($p>0.05$) (Table 3).

In this study, the parasitological examinations revealed various infections caused by different parasitic helminths. Isolated types of helminths were found in the digestive tracts and gills of both *Sarpa salpa* and *Euthynnus alletteratus*, classified at the class or order level. The study identified helminths from the Phylum Platyhelminths, specifically Class Trematoda, including Monogenea and the bi-subclass Digenea, in both fish species. Additionally, helminths from the phylum Acanthocephala were also isolated.

The morphological and morphometric analysis of various helminth species collected from *Euthynnus alletteratus* indicated that Monogenea were the most prevalent parasites, accounting for 66.7%, followed by Digenea at 33.3%, both collected from the gills. Additionally, Digenea collected from the digestive tract represented 66.7% of the total infections in *Euthynnus alletteratus*.

Monogenea, found in the gills, are hermaphroditic helminths with simple life cycles. Most of these parasites inhabit the external parts of marine fish, with a few residing in the digestive tract. They primarily feed on the mucous membranes of the gills and can be pathogenic, often leading to secondary infections[18].

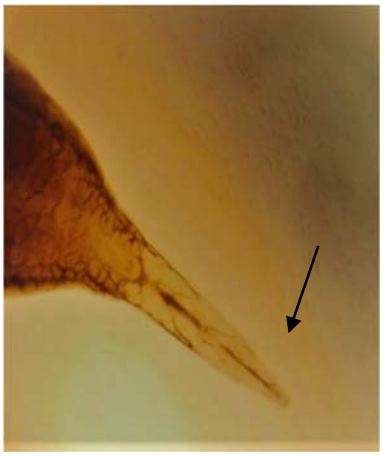

Fig. 1: External shape of Monogenea, with an arrow indicating the front part at a magnification of 10x
This detailed view highlights the morphological features characteristic of this group of helminths

Fig. 2: Exterior of Monogenea, with an arrow indicating the back part at a magnification of 10x
This illustration emphasizes the morphological traits essential for identifying this type of helminth

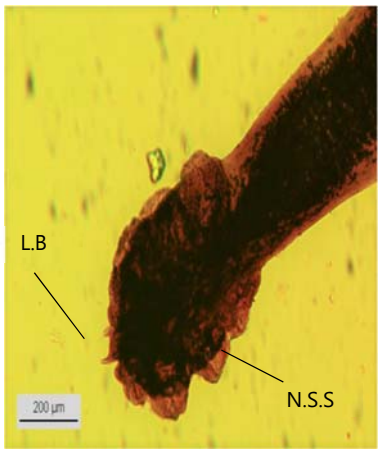

Fig. 3: Opisthaptor of the Monogenea (N.S.S), emphasising the number of small suckers and two little bits (L.B)
This image offers insight into the structure and attachment mechanisms of these helminths

The morphological characteristics of the Monogenea in this study revealed a strong posterior sucker with numerous small suckers equipped with fine spines, classifying them under the order Polyopisthocotylea[19] as illustrated in Fig. 1-4.

Fig. 4: Opisthaptor of the Monogenea, showing the number of small suckers at a magnification of 10x
This detailed view highlights the structural adaptations that facilitate their attachment to hosts

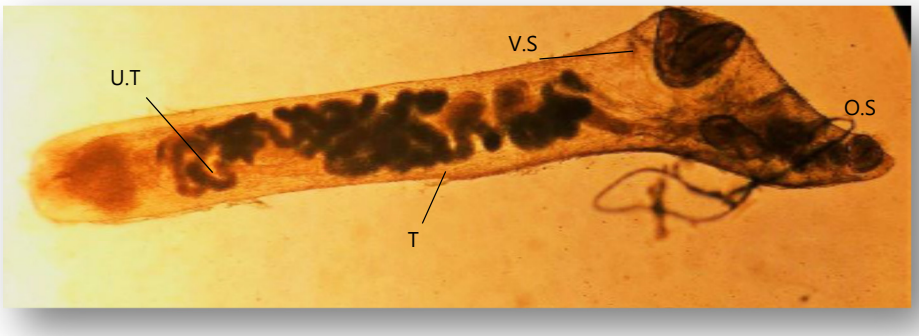

Fig. 5: Exterior of Digenea, labelled with the following: S.S.: Oral sucker, V.S.: Ventral sucker, T: Testes, and U.T.: Uterus, all at a magnification of 10x
This image highlights the key morphological features of Digenea

Regarding the Digenea collected from the gills and digestive tract, these helminths are hermaphroditic with complex life cycles. Most are parasitic in the internal organs of marine fish, while some infect the gills. They feed on the connective tissues of the gills and can be pathogenic to fish[18]. The morphological features of this digenean include a small oral sucker, a larger ventral sucker, and spherical testicles located above the egg-filled uterus, classifying them under the order[19] Azygiida as shown in Fig. 5.

The Digenea collected from the digestive tract feed on the mucous membranes lining the stomach and intestinal tract and are often pathogenic to fish[18]. Morphologically, this digenean is characterized by two suckers: The oral sucker surrounding the mouth opening and a larger abdominal sucker located in the middle of the body. Additionally, there is a pair of alternating testicles situated above the uterus. These helminths belong to the order Azygiida[19], as illustrated in Fig. 6-7.

On the other hand, the results of our study revealed that no cestode infections were detected in the examined fish. However, Acanthocephala infections were present, accounting for 33.3% of the total helminth infections observed. Based on the morphological and morphometric characteristics of the helminths recovered from the digestive tract of *Euthynnus alletteratus*, the parasites were identified as belonging to the phylum Acanthocephala. This group is distinguished by the presence of tentacles armed with fine spines, a complex life cycle, and their propensity to infect the digestive systems of their hosts. These helminths possess a long, double-walled proboscis covered with molecular spines, and their outer cuticle is lined with sharp spines that facilitate attachment to host tissues. According to Bursey *et al.*[20], these features classify them within the order Palaeacanthocephala, as illustrated in Fig. 8-9.

Fig. 6: Exterior of Digenea isolated from the alimentary duct of *Sarpa salpa*, captured at a magnification of 10x
This image highlights the morphological features specific to this helminth

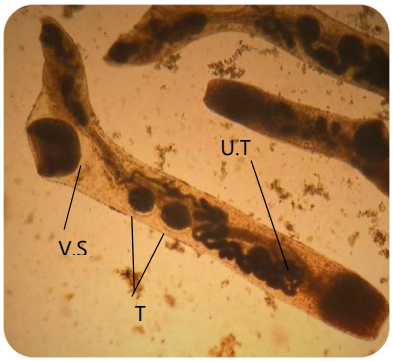

Fig. 7: External form of a digenean worm isolated from the alimentary tract of Sarpa salpa, with the following labels: V.S. for the ventral sucker, T for the testes, and U.T. for the uterus, all at a magnification of 10x
This image emphasises the key morphological characteristics of the helminth

Fig. 8: General view of an Acanthocephalan parasite isolated from the digestive tract of *Euthynnus alletteratus*, observed under 10× magnification

Based on the morphological and morphometric characteristics of helminths collected from various regions of *Sarpa salpa*, the most prevalent parasites were digeneans, accounting for 70% of the infections, and were primarily isolated from the digestive tract. In contrast, monogeneans were collected from the gills. Morphological examination of these helminths revealed the presence of a prominent posterior attachment organ consisting of a single robust sucker equipped with strong hooks or claws. These defining features are characteristic of worms belonging to the order Monopisthocotylea, as described[21-23], and are illustrated in Fig. 10.

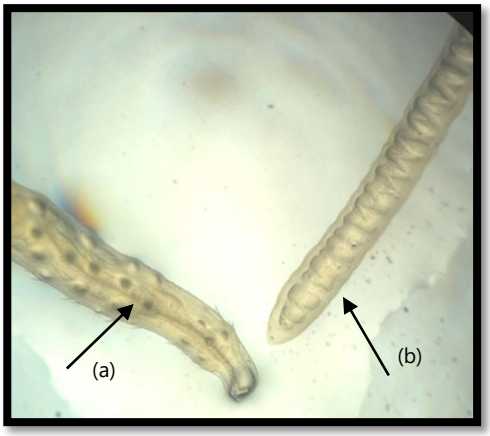

Fig. 9: Morphological features of an Acanthocephalan parasite isolated from the digestive tract of *Euthynnus alletteratus*, (a) Head region exhibiting fine spines (indicated by arrows), and (b) posterior end
Images captured under magnification of 40X

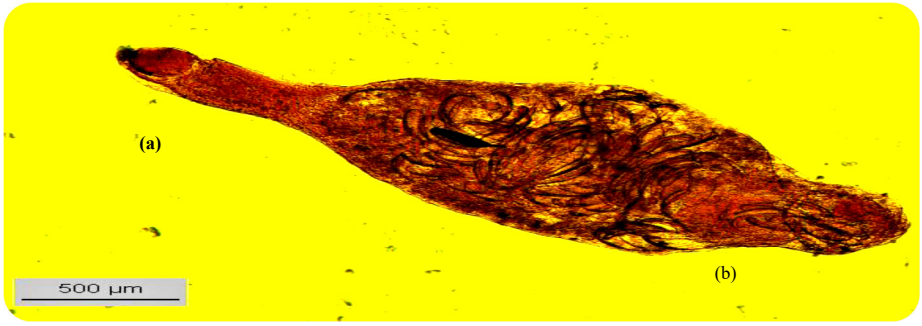

Fig. 10: General view of a Monogenean parasite isolated from the gills of *Sarpa salpa*, (a) Anterior region of the worm's body and (b) Posterior region showing a single haptor (sucker), characteristic of members of the order Monopisthocotylea

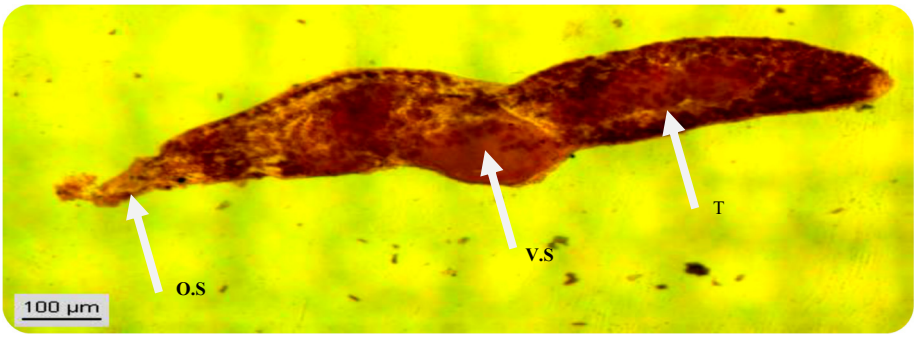

Fig. 11: General view of digeneans isolated from the alimentary tract of *Sarpa salpa*. Key anatomical features are labeled, including the O.S: Oral sucker, V.S: Ventral sucker and T: Testes

In this study, the digenean parasite isolated from the digestive tract exhibited distinct morphological features. The anterior end of the worm is tapered and bears a small oral sucker, while a larger ventral sucker is located mid-body. A follicular sac is positioned above the ventral sucker, and the testes are spherical and situated at the posterior end of the body. Based on these characteristics, the parasite is classified within the order Plagiorchiida[24], as illustrated in Fig. 11.

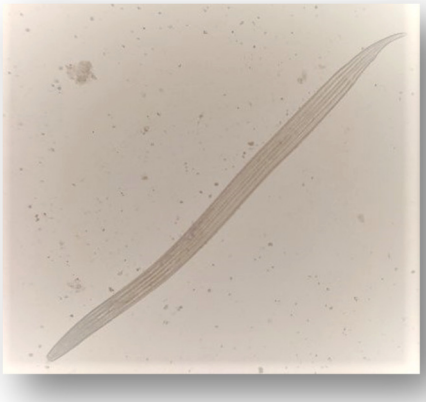

Fig. 12: Light micrograph of a nematode larva (order Rhabditida) isolated from the intestinal tract of *Sarpa salpa*, 10× magnification
Note the lip-like anterior structures (arrow) and coiled body wall

Fig. 13: Light micrograph of a filamentous nematode larva (Rhabditida) from the intestinal tract of *Sarpa salpa* (10×)
Note the slender, elongated body and undulating cuticle (arrows)

Fig. 14: Anterior morphology of a Rhabditida larva from *Sarpa salpa* intestine, highlighting the terminal mouth opening (arrow) flanked by three distinct lip-like papillae

During this study, no cestode infections were detected in *Sarpa salpa*. However, nematode infections were observed in the digestive tract at a prevalence rate of 30%. These helminths belong to the phylum Nemathelminthes and the class Nematoda (Fig. 12-14). The nematodes were slender, with anterior lip-like structures, and were primarily found in their larval stages. They exhibited an irregular, coiled body wall and a simple oesophagus. Based on morphological characteristics, these worms were identified as belonging to the order Rhabditida[25].

In this study, 140 bony fish specimens-70 *Sarpa salpa* (Shalaba) and 70 *Euthynnus alletteratus* (Razam)-were collected from the Qasr Ahmed local market for parasitological examination. The weights of *Sarpa salpa* ranged from 235-350 g, while *Euthynnus alletteratus* weighed between 840-1320 g.

Analysis revealed no correlation between fish weight and the presence of parasitic infection. However, a relationship was observed between weight and infection severity, with lighter fish exhibiting higher parasitic burdens. This trend may be attributed to environmental factors or stochastic chance, consistent with findings by Britton *et al.*[15]. Among the total fish examined in our study, 100 (71.4%) were found to be infected with Trematoda, Monogenea, and Acanthocephala, collected from the digestive tract and gills. Of these infected fish, 7 (70%) were *Euthynnus alletteratus* naturally infected with helminth parasites, while only 3 (30%) were *Sarpa salpa*.

Regarding parasite burden, a total of 1,308 helminths were isolated from the examined fish, with *Euthynnus alletteratus* harboring the majority (1,258 parasites, 96.2%) compared to *Sarpa salpa* (50 parasites, 3.8%). This higher prevalence in *Euthynnus alletteratus* aligns with findings from Italian coastal waters, where this species also showed elevated infection rates in both gills and digestive tracts[16].

Parasitological examination revealed distinct helminth communities in two fish species. In *Euthynnus alletteratus*, Monogenea predominantly caused gill infections, accounting for 66.7% of the infections, while gastrointestinal tract infections were primarily due to Digenea, also at 66.7%. The presence of this Digenean aligns with findings from Taiwan regarding a related *Euthynnus* species[26]. The prevalence rate of Monogenea was similar to studies on the same host in the Mediterranean Basin, such as those conducted in Italy (55%[26]) and Tunisia[27], suggesting that this parasite is endemic to the region. However, these rates were lower than those reported for *E. alletteratus* in Brazil[28], which may be due to geographic variation. Notably, no cestode infections were found in *E. alletteratus*. However, *Acanthocephala* infections were present at a rate of 33.3%, a finding consistent with results from Indonesia for a related species (26.7%[29]).

In *Sarpa salpa*, a single Monogenean (order: Monopisthocotylea) was isolated from the gills. This genus has previously been recorded in Egypt in a fish species of the same family[30]. Digeneans were the most common parasites in *S. salpa,* constituting 70% of infections and found in the digestive tract. This prevalence is lower than the 93.8% reported in a study conducted in Algeria[31], and the 10% reported by Gargouri Ben Abdallah *et al.*[6], but similar to findings in Turkey[32]. These variations may be attributed to differences in study areas or sampling seasons.

No cestodes were found in *S. salpa*, but nematodes (order: Rhabditida) were present at a prevalence of 30%. This contrasts with a study in Turkey that reported no nematode larvae in *S. salpa*[25]. This discrepancy is likely due to differences in the aquatic environments between Libya and Turkey, as well as dietary factors since nematode larvae are often transmitted through the consumption of intermediate hosts[33].

CONCLUSION

Fish parasites significantly contribute to economic losses in fisheries. This study highlighted a notable difference in the severity of helminth infections between two species: The carnivorous *Euthynnus alletteratus* (Razam) and the herbivorous *Sarpa salpa* (shalaba). The Razam fish, which feeds on crustaceans and small fish, exhibited a significantly higher intensity of parasitic helminth infections. This is likely because many parasitic worms have complex life cycles that rely on intermediate hosts, such as the prey consumed by carnivorous fish. Additionally, the high severity of infections is influenced by the morphological differences in the digestive tracts of teleosts and the specific ecological preferences of the parasites. In contrast, the herbivorous Shalaba fish showed a much lower severity of infection. This

difference can be directly attributed to their distinct diets and trophic levels. Therefore, it is crucial to develop strategies to mitigate helminth parasite infections in fish, particularly in commercially valuable carnivorous species, to reduce the associated economic impacts.

SIGNIFICANCE STATEMENT

This study provides baseline evidence on helminth parasite diversity, prevalence, and tissue damage in two marine fish species with contrasting feeding habits in the Misurata Region. The findings highlight the role of diet in shaping infection patterns and parasite burden. These results contribute to understanding fish health, parasite transmission dynamics, and may support improved monitoring and management of marine fisheries resources.

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