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Histopathological Changes Caused by Parasites in *Carangoides Bajad* Fish in the Red Sea, Jeddah

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RESEARCH ARTICLE

Abstract

This research focused on discovering parasites in the fish *Carangoides bajad* (orange-spotted trevally) (Forsskal, 1775) that were captured from the Red Sea off Saudi Arabia. *Carangoides bajad* (n = 120) were collected during the spring and summer of 2019. Internal parasites were extracted and subjected to parasitological analyses and fish intestines were subjected to histopathological analyses. The average length and weight of *Carangoides bajad* were 39.88 ± 8.7 cm and 1.68 ± 0.58 kg, respectively. We identified *Anisakis* (17.5%), *Lecithocladium* (23.33%), and *Bucephalus* spp (29.17%) in the infected fish. Female fish outnumbered males, suggesting that sex significantly influence the prevalence of parasitic infections. Parasites, or evidence of parasitic infection, were not detected in small fish (weighing 0.5–0.90 kg or 1.0–1.49 kg, and 20.0–29.9-cm long). Similarly, the values of Fulton's condition factor (*K*) ranged between 3.0–3.99 and 4.0–4.99, supporting the conclusion that these small fish were not infected with parasites. The data on larger fish (weighing 1.5–1.90 kg or 2.0–2.49 kg or 2.5–2.99 kg and length 20.0–29.9 cm or 30.0–39.9 cm or 40.0–49.9 cm or 50.0–59.9 cm) reveal that parasitic infections significantly influenced the length, weight, and Fulton's condition factor (*K*) according to the prevalence and intensity of infection of *Carangoides bajad*. Histopathological examination revealed intestinal tissue damage; and 45% of inflamed tissues involved swelling of the intestinal villi. Further, we observed separation of the mucosal epithelia from the submucosa, proliferation of goblet cells, and lymphocyte infiltration accompanied by atrophy and lysis of intestinal villi. The present study demonstrates infection of *Carangoides bajad* with three types of parasites and documents their deleterious effects.

Keywords: *Carangoides bajad*; *Anisakis* sp.; *Lecithocladium sp.; Bucephalus sp.;* Histopathology.

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INTRODUCTION

Parasites are ubiquitously and globally distributed, such that parasite-free environments are difficult to find. Parasites live in a dynamic equilibrium with their host. However, changes in the balance between the life cycle of a parasite and its host may lead to death of the latter. Such changes may involve the climate or human activities, including pollution and urbanization (Jones et al., 2008; D'Odorico et al., 2014). Changes within the host caused by parasitic infection may be detrimental, and it is therefore important to understand the characteristics of parasites within a given community. Diseases caused by specific environmental and societal conditions require a strategy to reduce their risks. It is therefore important for researchers to constantly consider the effects of parasites on the health of fish, the local community, and the associated ecosystem. Widespread parasitic infections are major causes of fish mortality. *C. bajad* (Forsskal, 1775) (Carangoidae), a demersal crangid, is a populous (Bhure et al., 2016) and economically important species that inhabits the Red Sea (Al-Zubaidy, 2010). *C. bajad* feeds on invertebrates such as crustaceans and planktons. Parasites have a major impact on the life cycle of their host (Al-Zubaidy, 2010). Protozoa and monogenetic trematodes are parasites that cause significant damage to fish gills,

resulting in anemia and death of the host (Morsy et al., 2014). Nematodes, cestodes, and trematodes cause human zoonoses, such as trichinellosis caused by roundworms (schistosomiasis) (Robinson and Dalton, 2009). Helminthic infections of humans, particularly those caused by parasitic worms, are a primary concern because many species are significant human pathogens (Garcia et al., 2007; Robinson and Dalton, 2009). There are many essential reasons that fish that inhabit fresh and brackish waters sustain human trematode infections, as indicated by the growing body of knowledge related to the variety and prevalence of food-borne trematodes present in Asian nations (Khalil et al., 2014).

The decades-long, industry-wide issue of managing diseases, parasites, and pests persists; and the impact of climate change on aquaculture is unknown and impossible to predict (Naylor et al., 2021). Further, recent outbreaks of emerging illnesses such as white spot, acute hepatopancreatic necrosis, and microsporidian parasites have caused notable losses in production, which have a significant economic impact (Robinson and Dalton, 2009). Parasites and bacteria play a normally limited role, although when animals are confined and subjected to stress, the impacts of these organisms may be significant.

Culture conditions, including increased nutritional levels, lead to an increase in the growth of parasitic populations (Amos et al., 2018). Although marine fish are susceptible to helminthic parasitic infections, the prevalence of parasitism is likely universal (Rio Indaryanto et al., 2015). Organisms such as Anisakis are the only parasites in seafood that may cause allergic responses (EFSA, 2010). These parasites most frequently populate the Atlantic, Pacific, and Mediterranean areas, causing anisakiasis etiologically associated with poor quality fish products (Buchmann and Mehrdana, 2016; Bao et al., 2017). The present study focused on detecting internal parasites among *C. bajad* inhabiting the Red Sea area of Jeddah and analyzing the histopathological changes caused by species representing three genera of parasites.

MATERIALS AND METHODS

Study area

This study area was located in the Red Sea, Jeddah, south of Jeddah City, approximately 5,400 km from the coast (longitude 21°28°37'N and latitude 39°11°49'E) (Figure 1). The study area was chosen because of its proximity to the Saudi Aramco Refinery, the Jeddah Islamic Seaport, and sources of industrial sewage waste to the south.

Fish samples

C. bajad specimens (n = 120) were captured using fishing cages and fishing rods in five locations around the study area. The areas are separated from each other by at least 500 m. The catch was harvested twice each month for three months during the spring and summer of 2019. *C. bajad* was chosen for its economic and commercial importance, attributable to its palatable and unique taste. Morphometric data included total weight (kg) and total length (cm), and Fulton's condition factor (*K*) was determined using a measuring tape and a balance (Mojekwu and Anumuda, 2015).

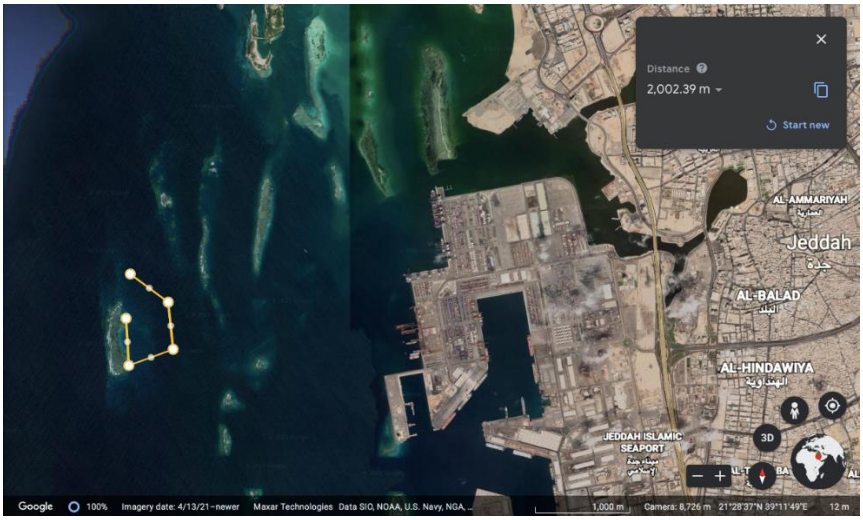

Figure 1: Study area

Parasitological examination

After dissection, the fish were inspected for internal parasites and placed in physiological saline solution. The intestines were dissected and portions were placed on a clean petri dish. Each segment was longitudinally opened, the mucosal layer was scraped from its wall with a blunt scalpel, and the contents were collected in a glass cup

containing a physiological saline solution. The glass cups were left for approximately 1–2 h until the parasites detached from the intestinal wall. The contents were filtered, and the collected parasites were washed several times with a physiological saline solution and filtered again to separate sediment or mucous membranes. The solution was then distributed among several dishes for examination using a light microscope.

Histological examination

Dissected intestinal tissues were examined for histological changes, submerged in a neutral formalin solution for 24 h, dried, and submerged in wax. A 2–3-µ section was stained with Hematoxylin and Eosin (H&E). The histological damage to intestinal tissues was evaluated and assigned a Lesion Score (LS). Semi-quantitative histological evaluation was performed according to Kocan et al. (1998) through allocating the histological damage according to severity into the groups as follows: None, Scarce, Moderate, and Severe. Tissue sections of each fish were analyzed to identify damage to tissues and cells. The histopathological damages in each fish were noted and their percentage prevalence was estimated by using the formula as mentioned below:

$$Percentage\ of\ histopathological\ damage\ in\ fish = \left(\frac{Total\ number\ of\ fish\ inspected\ with\ each\ damage}{total\ number\ of\ fish\ analyzed}\right) \times 100$$

Statistical analysis

Data analysis was performed using SAS for Windows (version 8.2) (SAS Institute Inc., Cary, NC USA). Data are expressed as the mean ± standard deviation. The prevalence and mean intensity of parasite infection in collected fish was calculated using SAS for Windows (version 8.2) (SAS Institute Inc., Cary, NC USA). Initially, descriptive statistical analysis was employed for analysis of data obtained. The percentage prevalence of infection was determined as percentage of the total number of fish infected out of total number of fish examined. Mean intensity was expressed as ratio between the number of parasite and number of examined fish. Since the data was nominal without normal distribution, the significance of difference was calculated using chi-square test. Variation in prevalence, mean intensity with respect to length, weight and *K* of fish was statistically analyzed using one-way ANOVA with spearman test (Rho). In addition, dependence of infection on sex of the fish was analyzed by t-test. The following hypothesis was assumed for the study,

H_0 (null hypothesis): There is no statistical significance of length, weight and *K* of fish on parasite prevalence.

H_1 (alternative hypothesis): There is statistical significance of length, weight and *K* of fish on parasite prevalence.

The result of the analysis when showing more than 0.05 significance indicate that the alternative hypothesis can be accepted. When the test results are significant, post-hoc power analysis indicated statistical power >0.08, as described by Britton et al. (2011).

RESULTS AND DISCUSSIONS

Biometric data

The average length and weight of the 120 *C. bajad* harvested from the waters off Jeddah were 39.88 ± 8.7 cm and 1.68 ± 0.58 kg, respectively. The value of *K* (2.77 ± 0.84) indicated that body weights indicated good health. Skewness and kurtosis values indicated that the data were symmetric and normally distributed (Table 1).

Table 1: Descriptive statistics of biometric data of *C. bajad*

Parameters	Mean	Standard error of mean	Standard Deviation	Variance	Skewness	Kurtosis
Weight	1.68	0.05	0.58	0.34	−0.22	−1.00
Length	39.88	0.79	8.70	75.64	−0.04	−1.04
Fulton's condition factor	2.77	0.08	0.84	0.71	0.37	−0.99

Table 2 shows that the prevalence of three species of parasites in female and male *C. bajad* was 70% (84/120). Intestinal parasites identified according to their morphological features were as follows: *Anisakis* (no. of fish infected = 21, 17.5%), *Lecithocladium* (no. of fish infected = 28, 23.3%), and *Bucephalus* spp. (no. of fish infected = 35, 29.2%) (Figure 2). Totally, 35 *Anisakis* sp., 35 *Lecithocladium* sp. and 70 *Bucephalus* sp. were isolated from 84 specimens. *Bucephalus* sp. was most common with 29.17% prevalence, followed by *Lecithocladium* sp. (23.33%).

Anisakis sp. was found in minority (17.50%). Further, investigation was conducted to determine the differences in the prevalence of parasitic infection in male and female fishes (H_0: Gender had no influence on parasitic prevalence). The prevalence of infected female fish with *Anisakis* sp. (66.66%) was significantly higher compared with that of males (33.33%) (χ^2 = 2.82, p = 0.09), Similarly, significant difference was noticed between the percentages of infected female (75%) and male (25%) with *Lecithocladium* sp. (χ^2 = 2.43, p = 0.29), as well as between infected female and male with *Bucephalus* sp. (female 80% and male 20%) (χ^2 = 0.00, p = 1.00). The obtained data rejected the null hypothesis and indicates gender showed significant difference.

Table 2: Prevalence and intensity of infection according to sex

Type of parasite	Variables	Number infected	Prevalence (%)	Number of parasites recovered	Intensity	Odds ratio (95% CI)
Anisakis sp.	Male (n = 24)	7	33.33	12	1.71	0.415 (0.146–1.181)
	Female (n = 96)	14	66.66	23	1.64	
	Chi-square	-	2.82	-	6.21	
	p-value	-	0.09	-	0.10	
	Total (n = 120)	21	17.50	35	1.67	
Lecithocladium sp.	Male (n = 24)	7	25.0	10	1.43	0.68 (0.249–1.857)
	Female (n = 96)	21	75.0	25	1.19	
	Chi-square	-	0.57	-	2.43	
	p-value	-	0.45	-	0.29	
	Total (n = 120)	28	23.33	35	1.25	
Bucephalus sp.	Male (n = 24)	7	20.0	14	2.00	1 (0.374–2.675)
	Female (n = 96)	28	80.0	56	2.00	
	Chi-square	-	0.00	-	4.12	
	p-value	-	1.00	-	0.25	
	Total (n = 120)	35	29.17	70	2.00	

- not applicable

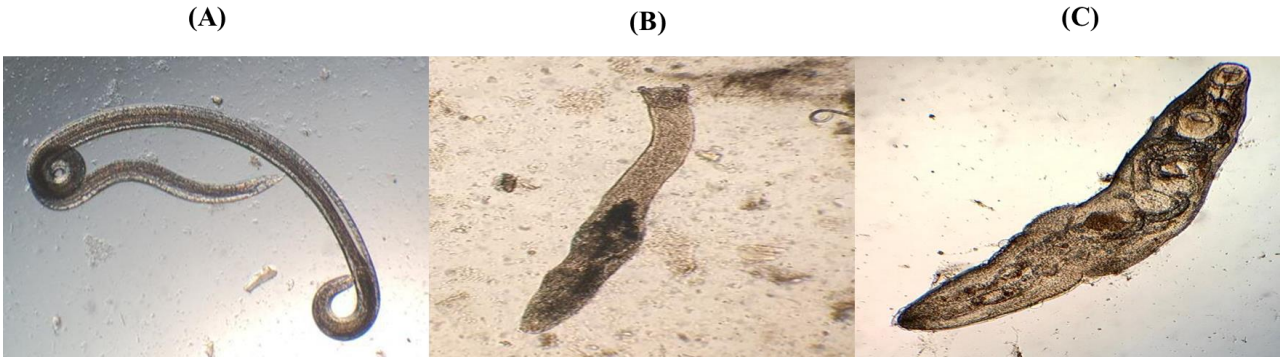

Figure 2. *Anisakis sp*. larvae (A), *Bucephalus sp*. (B), and Le*cithocladium sp* (C). in the intestines of *C. bajad* (Forsskål, 1775).

Prevalence and intensity of Anisakis sp. infection among C. bajad

Table 3 presents the prevalence and intensity of *Anisakis* sp. infection according to these three variables. (H_0: Length of fish had no significance on the prevalence and intensity of *Anisakis* sp). The lengths of the fish were allocated into two major ranges as follows: 40–49.9 cm (n = 45) and 50–59.9 cm (n = 15). *Anisakis* sp. infection, which was not prevalent in smaller fish (20–29.9 cm long), significantly increased as a function of length of their host (66.67%, 50–59.9 cm). Chi-square test (χ^2 = 36.3, p = 0.00) and mean intensity (χ^2 = 39.17, p = 0.00) reveal significant influence of length ($p < 0.05$) in the prevalence of *Anisakis* sp.

Table 3. Prevalence and intensity of infection of *Anisakis sp.* associated with length, weight, and Fulton's condition factor

Parameters	Variables	No. infected	Prevalence (%)	No. of recovered parasites	Intensity
Length	20.0-29.9 (n = 22)	0	0	0	0
	30.0-39.9 (n = 38)	1	2.63	2	2
	40.0-49.9 (n = 45)	10	22.22	17	1.7
	50.0-59.9 (n = 15)	10	66.67	16	1.6
	Chi-square		36.3		39.17
	p-value		0.00		0.00
	Total	21	17.5	35	1.67
Weight	0.5-0.90 (n = 23)	0	0.00	0	0.00
	1.0-1.49 (n = 19)	0	0.00	0	0.00
	1.5-1.90 (n = 36)	2	5.56	3	1.50
	2.0-2.49 (n = 34)	14	41.18	24	1.71
	2.5-2.99 (n = 8)	5	62.50	8	1.60
	Chi-square	-	36.90	-	38.30
	p-value	-	0.00	-	0.00
	Total	21	17.50	35	1.67
Fulton's Condition (k) Factor	1.0-1.99 (n = 27)	16	59.26	24	1.5
	2.0-2.99 (n = 47)	5	10.64	11	2.2
	3.0-3.99 (n = 35)	0	0	0	0
	4.0-4.99 (n = 11)	0	0	0	0
	Chi-square	-	43.9	-	51.9
	p-value	-	0.00	-	0.00
	Total	21	17.5	35	1.67

- not applicable

The weights of the fish, which were allocated into five groups, ranged from 0.5 kg to 3.0 kg (H_0: weight of fish does not influence prevalence and incident of *Anisakis* sp.) (Table 3). The majority of fish (n = 36) weighed between 1.5 kg to 1.9 kg and the minority (n = 8) weighed between 2.5 kg to 2.99 kg. All fish infected with *Anisakis* sp. weighed between 1.5 kg to 2.99 kg, indicating that uninfected fish do not support intestinal infection caused by *Anisakis* sp.

The maximum prevalence (62.50%) of infection was associated with fishes weighing between 2.5 kg to 2.99 kg., and there was a significant association between weight and infection with *Anisakis* sp. Chi-square test ($\chi^2 = 36.9$, p = 0.00) and the mean intensity ($\chi^2 = 38.3$, p = 0.00) reveal that there was a significant association between weight and infection with *Anisakis* sp. ($p<0.05$).

Fishes were allocated into four groups according to *K* as follows: (H_0: *K* factor had no influence on prevalence and intensity of *Anisakis* sp.) The maximum number of all fish (47/120) had *K* values ranging between 2 and 2.99. The highest number of infected fish (n = 16) had *K* values ranging between 1 to 1.99. The *K* values significantly decreased as a function of infection score. For example, fish with *K* values ranging from 1–1.99 had the highest prevalence (59.26%) of *Anisakis* sp (24 parasites recovered with type 2 error less than 0.2 and power over 0.8). Chi-square test ($\chi^2 = 43.9$, p = 0.00) and mean intensity ($\chi^2 = 51.9$, p = 0.00) reveal significant influence of *K* factor ($p<0.05$) in the prevalence and intensity of infection with *Anisakis* sp.

Prevalence and intensity of Lecithocladium sp. infection among C. bajad

Table 4 presents data for the prevalence and intensity of *Lecithocladium* sp. infection in *C. bajad* according to length, weight, and *K*. A total of 35 *Lecithocladium* sp. were recovered out of 120 samples analyzed. The fish were allocated into four groups according to length (H_0: Length of fish do not influence on *Lecithocladium* sp. prevalence). Among the 120 specimens, the lengths of 45 ranged between 40–49.9 cm, and the fewest (n = 15) were 50–59.9 cm long. The prevalence rate varied as a function of length. Thus, the lengths of 15 of 28 fish harboring 19 parasites ranged between 40–49.9 cm. The maximum prevalence (80%) of *Lecithocladium* sp. was associated with fish

harboring 14 parasites and ranging in length between 50 cm–59.9 cm. Chi-square test (χ^2 = 45.20, p = 0.00) and mean intensity (χ^2 = 47.13, p = 0.00) reveal significant association of length (p<0.05) in the prevalence of *Lecithocladium* sp.

Table 4. Prevalence and intensity of infection of *Lecithocladium sp.* associated with length, weight, and Fulton's condition factor

Parameters	Variables	Number infected	Prevalence (%)	Number of parasites recovered	Intensity
Length	20.0-29.9 (n = 22)	2	9.09	0	0.00
	30.0-39.9 (n = 38)	1	2.63	2	2.00
	40.0-49.9 (n = 45)	15	33.33	19	1.27
	50.0-59.9 (n = 15)	12	80.00	14	1.17
	Chi-square	-	45.20	-	47.13
	p-value	-	0.00	-	0.00
	Total	28	23.33	35	1.25
Weight	0.5-0.90 (n = 23)	0	0.00	0	0.00
	1.0-1.49 (n = 19)	0	0.00	0	0.00
	1.5-1.90 (n = 36)	4	11.11	5	1.25
	2.0-2.49 (n = 34)	19	55.88	24	1.26
	2.5-2.99 (n = 8)	5	62.50	6	1.20
	Chi-square	-	42.80	-	43.00
	p-value	-	0.00	-	0.00
	Total	28	23.33	35	1.25
Fulton's Condition Factor	1.0-1.99 (n = 27)	20	74.07	24	1.20
	2.0-2.99 (n = 47)	8	17.02	11	1.38
	3.0-3.99 (n = 35)	0	0.00	0	0.00
	4.0-4.99 (n = 11)	0	0.00	0	0.00
	Chi-square	-	53.91	-	55.24
	p-value	-	0.00	-	0.00
	Total	28	23.33	35	1.25

- not applicable

The fish were assigned to allocated into five categories according to weight (H_0: Prevalence and intensity of *Lecithocladium* sp. infection was not affected by the weight of the fish). The highest number of fish (n = 36) weighed 1.5 kg to 1.9 kg and the fewest (n = 8) weighed 2.5 kg to 2.99 kg. *Lecithocladium* sp. was not observed in fish weighing between 0.5 kg to 1.49 kg. The maximum prevalence (62.5%) was for fish weighing 2.5 kg to 2.99 kg. Chi-square test (χ^2 = 42.8, p = 0.00) and mean intensity (χ^2 = 43.0, p = 0.00) reveal significant influence of weight (p< 0.05) in the prevalence of *Lecithocladium* sp. These findings indicate a significant influence of weight on the prevalence of *Lecithocladium* sp. Similar to the prevalence of *Anisakis* sp., that of *Lecithocladium* sp. decreased in association with increasing *K* values. Thus, *K* values ranging between 1 to 1.99 were associated with the highest prevalence of infection (74.07%); and 24 parasites were recovered from this weight group. Chi-square test (χ^2 = 53.91, p = 0.00) and mean intensity (χ^2 = 55.24, p = 0.00) reveal significant influence of *K* on the prevalence of *Lecithocladium* sp. and suggest that length, weight, and *K* influenced the prevalence and intensity of infection (with type 2 error less than 0.2 and power more than 0.8).

Prevalence and intensity of Bucephalus sp. infection among C. bajad

Table 5 presents data for the prevalence and intensity of *Bucephalus* sp. infection according to length, weight, and *K* (H_0: Length of fish had no influence on the prevalence and intensity of *Bucephalus* sp. infection). We recovered 70 *Bucephalus* sp. from the 35 specimens. The prevalence of *Bucephalus* sp. increased as a function of increasing length. Thus, parasitic infection was not detected in small fish, and the highest prevalence (86.67%) was observed

in fish ranging from 50 cm to 59.9 cm in length. Chi-square test (χ^2 = 52.69, p = 0.00) and mean intensity (χ^2 = 61.17, p = 0.00) reveal significant influence of length (p<0.05) in the prevalence of *Bucephalus* sp.

Table 5. Prevalence and intensity of infection *of Bucephalus sp.* associated with length, weight, and Fulton's condition factor

Parameters	Variables	Number infected	Prevalence (%)	Number of parasites recovered	Intensity
Length	20.0–29.9 (n = 22)	0	0.00	0	0.00
	30.0–39.9 (n = 38)	1	2.63	3	3.00
	40.0–49.9 (n = 45)	21	46.67	41	1.95
	50.0–59.9 (n = 15)	13	86.67	26	2.00
	Chi-square	-	52.69	-	61.17
	p-value	-	0.00	-	0.00
	Total	35	29.17	70	2.00
Weight	0.5–0.90 (n = 23)	0	0.00	0	0.00
	1.0–1.49 (n = 19)	0	0.00	0	0.00
	1.5–1.90 (n = 36)	7	19.44	17	2.43
	2.0–2.49 (n = 34)	23	67.65	42	1.83
	2.5–2.99 (n = 8)	5	62.50	11	2.20
	Chi-square	-	47.61	-	43.00
	p-value	-	0.00	-	0.00
	Total	35	29.17	70	2.00
Fulton's Condition Factor	1.0–1.99 (n = 27)	24	88.89	46	1.92
	2.0–2.99 (n = 47)	11	23.40	24	2.18
	3.0–3.99 (n = 35)	0	0.00	0	0.00
	4.0–4.99 (n = 11)	0	0.00	0	0.00
	Chi-square	-	66.30	-	67.96
	p-value	-	0.00	-	0.00
	Total	35	29.17	70	2.00

- not applicable

Similar to the association with length, *Bucephalus* sp. was not observed in small fish weighing between 0.5 kg to 1.49 kg (H_0: The weights of fish had no influence on the prevalence and intensity of Bucephalus sp. infection. The maximum prevalence of *Bucephalus* sp. was 67.65% in fish weighing between 2 kg to 2.49 kg, and this association was significant. Chi-square test (χ^2 = 47.61, p = 0.00) and mean intensity (χ^2 = 43.00, p = 0.00) reveal a significant influence of weight (p<0.05) in the prevalence of *Bucephalus* sp. Further, the highest prevalence (88.89%) was significantly associated with the group with *K* values ranging between 1 to 1.99. Thus, increasing *K* values were significantly associated with decreased prevalence of infection. For example, 46 parasites were recovered from fish with *K* values ranging between 1–1.99 (with type 2 error less than 0.2 and power more than 0.8). These data reveal a significant influence of *K* on the prevalence of *Bucephalus* sp. Chi-square test (χ^2 = 66.3, p = 0.00) and mean intensity (χ^2 = 67.96, p = 0.00) reveal significant influence of k-factor (p<0.05) in the prevalence of *Bucephalus* sp. Further, length, weight, and *K* exerted a great influence on the prevalence and intensity of *Bucephalus* sp. infection.

Histological changes caused by parasitic infection

Severe damage to intestinal tissue was observed, represented by swelling and adhesion of the intestinal villi caused by clear vacuolar degeneration in all layers of the intestine (Figure 3). The villi appeared atrophic, with disintegration, necrosis, and inflammatory cellular infiltration accompanied by an increase in muscular bundles (Plate 1). A localized increase in goblet mucosal cells was observed in some sections as well as vacuolar degeneration of mucosal epithelial cells, with disruption of muscular bundles and infiltration of lymphocytes (Plate 2). Many parasites were present in the intestinal lumen, along with sloughing of the intestinal mucosa, and its

inward separation with cellular infiltration and dead tissue (Plate 3). Most sections were characterized by the disrupted structural organization of the intestinal tissue, such as distorted and decomposed villi that were separated from the submucosal layer. Further, Plate 4 shows dense cellular infiltration of the intestinal tissues (Plate 4).

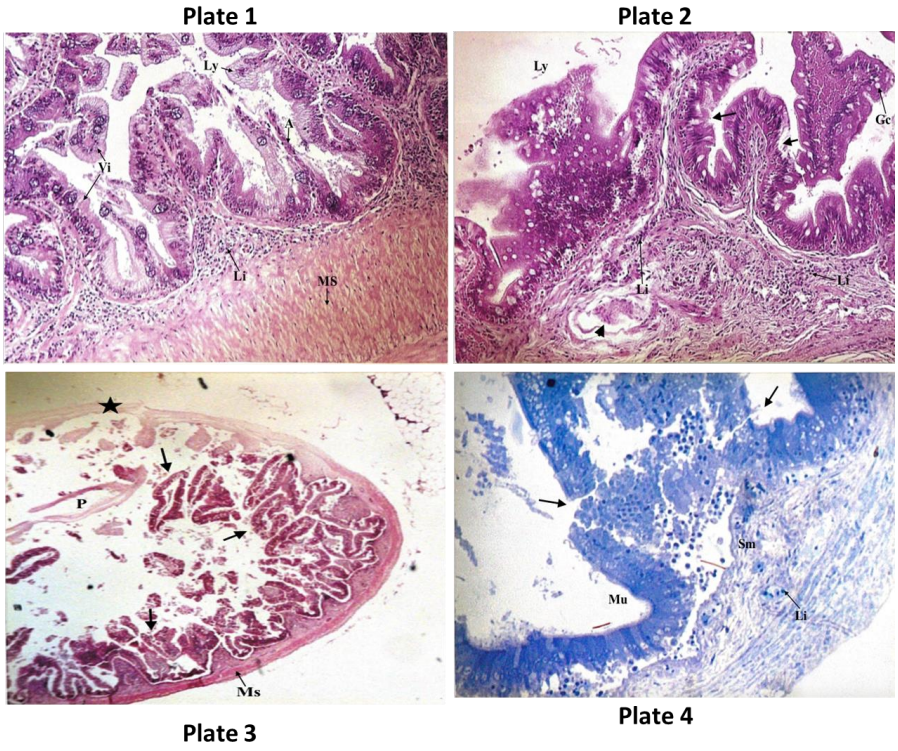

Figure 3. Histopathological changes in the intestines of *C. bajad* caused by parasitic infection. **Plate 1** shows atrophy (A); lysis (Ly), and lymphocytic infiltration (Li) in the submucosa (Sm), and increase in muscular bundles (Ms). Notice fusion and vacuolar degeneration in the intestinal villi. (H&E; Scale bar = 40 µm). **Plate 2** shows lysis of mucosal epithelial cells (Ly), lymphocytic infiltration (Li), and damaged muscular bundles (arrowhead). Notice goblet cells (Gc) and vacuolar and degeneration of mucosal epithelial cells (arrow); (H&E; Bar = 40 µm). **Plate 3** shows damage and sloughing of the intestinal villi (arrow); dead tissue (star); parasite in the Lumen (P); thinning of muscularis (Ms); (H&E; Bar = 200 µm). Plate 4 shows deformed villi, lysis of mucosal epithelial cells (Ly), separation of the mucosa (Mu) from the submucosa (Sm) and lymphocytic infiltration (Li). (H&E; Bar = 20 µm).

As detailed in the methods section, the infections were assigned scores according to the severity of inflammation (Table 6). For example, 93.33% of tissues were not infected. Parasitic infection was infrequent (5%) or Moderate (1.67%). Swelling of intestinal villi was high in moderate cases with 45% inflammation. Severe inflammation and damage to the intestines involved separation of mucosal epithelia from the submucosa (54.17%), proliferation of goblet cells (83.33%), and lymphocyte infiltration (41.67%). Atrophy and lysis of intestinal villi involved 33.33% of tissues. At the same time, damage and sloughing of mucosal epithelia and Atrophy and lysis of intestinal villi was high for Moderate score (45.83 and 33.33%, respectively). Fusion of the intestinal villi and loosening or reduction of the muscular bundles was noticed to be high for Scarce scoring with 35.83 and 33.33%, respectively. About 50.83% of parasites had not shown an extension of the intestinal lumen.

Table 6: Inflammatory scores of parasitic infections of *C. bajad*

Condition	None	Scarce	Moderate	Severe
Parasitic infection	**93.33**	5.00	1.67	0.00
Swelling of the intestinal villi	16.67	30.00	**45.00**	0.00
Separation of mucosal epithelia from sub mucosa	0.00	10.83	35.00	**54.17**
Proliferation of goblet cells	0.00	4.17	12.50	**83.33**
Lymphocytic infiltration	0.00	20.83	37.50	**41.67**

Damage and sloughing of mucosal epithelia	13.33	20.00	**45.83**	20.83
Extension of the intestinal lumen	**50.83**	18.33	15.00	15.83
Fusion of the intestinal villi	29.17	**35.83**	26.67	0.00
Atrophy and lysis of intestinal villi	20.83	25.00	**33.33**	20.83
Loosening or reduction of the muscular bundles	16.67	**33.33**	25.83	24.17

Here we show that longer specimens of *C. bajad* that inhabit the coastal waters of Jeddah experienced higher rates of parasitic infections compared with those of smaller fish. These findings indicate that older fish were likely exposed to parasites for longer periods. For example, Oniye et al. (2004) did not observe parasitic infections in young and small fish, although parasites are more prevalent in adult fish because of different diets. In the present study, *Bucephalus* sp. was numerous in *C. bajad*. However, the prevalence of *Bucephalus* sp. was higher in *C. bajad* ranging in length from 50.0–59.9 cm (86.67%) compared with those ranging in length from 40.0–49.9 cm (46.67). Similarly, older *C. bajad* ranging in size from 50.0–59.9 cm experienced infection with *Lecithocladium* sp. and *Anisakis* sp. at rates of 80.0% and 66.67%, respectively. These findings are consistent with those of *Lecithocladium angustiovum* from the Visakhapatnam Coast, India, which is present in 88.5% of parasitized fish (Madhavi & Lakshmi 2011). Others found that *L. angustiovum* parasitizes the stomach and intestines of 12.67% and 87.33%, respectively, of Indonesian short mackerels (Rio Indaryanto et al., 2015).

Anisakis sp. parasites are prevalent worldwide, frequently infecting mackerels, blue whiting, and the European and silver hake (Debenedetti et al., 2019). Anisakis (Anisakidae) and raphidascarid larvae (Raphidascaridae) are identified according to their morphological characteristics and are classified as Anisakis type I (Anisakidae) (33.65%) and *Hysterothylacium* spp. (Raphidascaridae) (11.59%) (Debenedeti et al., 2019). The overall parasitic population in silver hake is extremely low, and few species of the genus *Contracaecum* (Aniakidae) infect silver hake (Debenedeti et al., 2019). Here we found that the infection intensities of *Anisakis* and *Lecithocladium* spp. were lower compared with that of *Bucephalus* sp. However, more recent data available for *Bucephalus margaritae* only represent hosts other than carangids (Bray et al., 2019). However, several species of *Bucephalus* parasitize *C. bajad* in the Hodeidah area of the Red Sea Coast of Yemen (Al-Zubaidy, 2010).

The increase in the numbers of *Bucephalus* larvae in the viscera may contribute to their migration to the epidermal region. It was not feasible here to evaluate this possibility, because there were numerous larvae in these locations, which prevented establishing a significant association. Therefore, we suggest that *Bucephalus* migrates. To raise awareness, manufacturers must clearly explain the parasite and its effects, as well as the existence of parasitic larvae. The overall consequence is that parasites will spread worldwide and cause parasitic infection or allergic responses of humans as well as induce cross-reactivity with other invertebrate proteins such as those associated with *Anisakis* sp. (Aibinu et al., 2019).

Approximately 5 days after consuming an infected fish, gastrointestinal symptoms such as nausea, vomiting, and diarrhea may appear. Anisakiasis symptoms often take longer to appear in intestinal infection compared with gastric anisakiasis (Takabayashi et al., 2014). Several *Anisakis* larvae were recently discovered in raw fish, showing that potentially infected seafood may harbor a multitude of larvae, which may enter the gastrointestinal tract via different routes (Mizumura et al., 2018). Our present study of *C. bajad* shows that inflammation and lysis of the intestinal villi and the proliferation of goblet cells and lymphocyte infiltration causes more severe symptoms and mortality.

Parasites such as trematodes and nematodes may induce chronic granulomatous inflammation (Feist and Longshaw, 2008). Although not a common component of parasite-induced granulomas, rodlet cells proliferate during parasitic infections, causing serious illness (Reite, 2005). Compared with other studies (Mohammed et al., 2009; Bichi and Yelwa 2010), here we found a great prevalence of parasites in the intestine. Our present observations in this regard are consistent with those of others (Mohammed et al., 2009; Bichi and Yelwa 2010).
The incidence of parasitic infections may be explained by the availability of food and the feeding activities of the host. The physiological, morphological, and ecological aspects of host specificity contribute to species specificities of the parasites. Geographical distribution, local habitat, nutrition, and the nutritional sources of animals are the main ecological variables. Further, seasonal variations may restrict a parasite to a specific host (Bhure et al., 2016).

CONCLUSIONS

This study demonstrated through identifying the parasitic species that infect *C. bajad*, the risks posed by parasites to fish mortality. Larger fish are more susceptible to parasitic infections than smaller fish. Therefore, it is important to monitor the condition of the fish, as parasitic animals and histological examination are important tools for

diagnosing tissue damage. Therefore, the study recommends future research on other aspects of these parasites as indicators of changes in fish health and the environment.

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Conflicts of Interest
The authors declare that they do not have any conflict of interest.

REFERENCES

1. Aibinu IE, Smooker PM, Lopata AL. Anisakis Nematodes in Fish and Shellfish- from infection to allergies. Int J Parasitol Parasites Wildl. 2019; 9, 384–393. https://doi.org/10.1016/j.ijppaw.2019.04.007
2. Al-Zubaidy A. First Record of Lecithochirium sp. (Digenea: Hemiuridae) in the Marine Fish Carangoides bajad from the Red Sea, Coast of Yemen. J King Abdulaziz Univ Sci. 2010; 21, 85–94. https://doi.org/10.4197/mar.21-1.4
3. Amos SO, Eyiseh TE, Michael ET. Parasitic infection and prevalence in Clarias Gariepinus in Lake Gerio, Yola, Adamawa state. MOJ Anat Physiol. 2018; 5. https://doi.org/10.15406/mojap.2018.05.00229
4. Bao M, Pierce GJ, Pascual S, González-Muñoz M, Mattiucci S, Mladineo I, et al. Assessing the risk of an emerging zoonosis of worldwide concern: anisakiasis. Sci Rep. 2017; 7, 43699. https://doi.org/10.1038/srep43699
5. Bhure D, Nanwareb S, Jadhavc A. Prevalence and diversity of cestode parasites of freshwater fishes of genus *Channa Scopoli*, 1777. Wld Sci News. 2016; 33: 15–26.
6. Bichi A, Yelwa S. Incidence of piscine parasites on the gills and gastrointestinal tract of clarias gariepinus (teugels) at bagauda fish farm, Kkano. Bayero J Pure Appl Sci. 2010; 3. https://doi.org/10.4314/bajopas.v3i1.58732
7. Bray RA, Palm HW, Theisen S. Bucephalus damriyasai n. sp. (Digenea: Bucephalidae) from the blacktip trevally Caranx heberi (Bennett) (Perciformes: Carangidae) off Bali, Indonesia. Syst Parasitol. 2019; 96, 65–78. https://doi.org/10.1007/s11230-018-9828-7
8. Britton JR, Pegg J, Williams CF. Pathological and ecological host consequences of infection by an introduced fish parasite. PLoS One. 2011; 6, e26365–e26365. https://doi.org/10.1371/journal.pone.0026365
9. Buchmann K, Mehrdana F. Effects of anisakid nematodes Anisakis simplex (s.l.), Pseudoterranova decipiens (s.l.) and Contracaecum osculatum (s.l.) on fish and consumer health. Food Waterborne Parasitol. 2016; 4, 13–22. https://doi.org/10.1016/j.fawpar.2016.07.003
10. D'Odorico P, Carr JA, Laio F, Ridolfi L, Vandoni S. Feeding humanity through global food trade. Earth's Futur. 2014; 2, 458–469. https://doi.org/10.1002/2014ef000250
11. Debenedetti Á, Madrid E, Trelis M, Codes F, Gil-Gómez F, Sáez-Durán S, et al. Prevalence and Risk of Anisakid Larvae in Fresh Fish Frequently Consumed in Spain: An Overview. Fishes. 2019; 4, 13. https://doi.org/10.3390/fishes4010013
12. EFSA. Scientific Opinion on risk assessment of parasites in fishery products, EFSA J. 2010; 8. https://doi.org/10.2903/j.efsa.2010.1543
13. Feist SW, Longshaw M. Histopathology of fish parasite infections - importance for populations. J Fish Biol. 2008; 73, 2143–2160. https://doi.org/10.1111/j.1095-8649.2008.02060.x
14. Garcia HH, Moro PL, Schantz PM. Zoonotic helminth infections of humans: echinococcosis, cysticercosis and fascioliasis. Curr Opin Infect Dis. 2007; 20, 489–494. https://doi.org/10.1097/qco.0b013e3282a95e39
15. Jones KE, Patel NG, Levy MA, Storeygard A, Balk D, Gittleman JL, et al. Global trends in emerging infectious diseases. Nature. 2008; 451, 990–993. https://doi.org/10.1038/nature06536

16. Khalil MI, El-Shahawy IS, Abdelkader HS. Studies on some fish parasites of public health importance in the southern area of Saudi Arabia. Rev Bras Parasitol Veterinária. 2014; 23, 435–442. https://doi.org/10.1590/s1984-29612014082
17. Kocan R, Hershberger P, Winton J. Ichthyophoniasis: An Emerging Disease of Chinook Salmon in the Yukon River. J Aquat Anim Health. 2004; 16, 58–72. https://doi.org/10.1577/h03-068.1
18. Madhavi R, Triveni Lakshmi T. Metazoan parasites of the Indian mackerel, Rastrelliger kanagurta (Scombridae) of Visakhapatnam coast, Bay of Bengal. J Parasit Dis. 2011; 35, 66–74. https://doi.org/10.1007/s12639-011-0028-5
19. Mizumura N, Okumura S, Tsuchihashi H, Ogawa M, Kawasaki M. A Second Attack of Anisakis: Intestinal Anisakiasis Following Gastric Anisakiasis. ACG case reports J. 2018; 5, e65–e65. https://doi.org/10.14309/crj.2018.65
20. Mohammed A, Kalu AU, Yem IY et al. Bacteriological quality of *Clarias gariepinus* (Burchell, 1822) in Lake Alau, Borno State, Nigeria. Best J. 2009; 6(1): 15–18.
21. Mojekwu TO, Anumudu CI. Advanced techniques for morphometric analysis in Fish. J Aquac Res Development. 2015; 6: 8.
22. Morsy K, El Fayoumi H, Al Shahawy G, Fahmy M. *Diplectanum harid* sp. nov. and *Pseudohabdosynochus chlorostigma* sp. nov. (Monogenea: Diplectanidae) from Scaridae and Serranidae Hosts of the Red Sea. World J Fish Mar Sci. 2014; 6: 319-327.
23. Naylor RL, Hardy RW, Buschmann AH, Bush SR, Cao L, Klinger DH, et al. A 20-year retrospective review of global aquaculture. Nature. 2021; 591, 551–563. https://doi.org/10.1038/s41586-021-03308-6
24. Oniye SJ, Adebote DA, Ayanda OI. Helminth parasites of Clarias gariepinus (Teugels) in Zaria, Nigeria. J Aquat Sci. 2004; 19. https://doi.org/10.4314/jas.v19i2.20027
25. Reite OB. The rodlet cells of teleostean fish: their potential role in host defence in relation to the role of mast cells/eosinophilic granule cells. Fish Shellfish Immunol. 2005; 19, 253–267. https://doi.org/10.1016/j.fsi.2005.01.002
26. Rio Indaryanto F, Wardiatno Y, Tiuria R. Penyebaran Cacing Parasitik Pada Ikan Kembung Perempuan (Rastrelliger brachysoma) Di Pulau Jawa. J Perikan dan Kelaut. 2014; 4. https://doi.org/10.33512/jpk.v4i4.168
27. Robinson MW, Dalton JP. Zoonotic helminth infections with particular emphasis on fasciolosis and other trematodiases. Philos Trans R Soc Lond B Biol Sci. 2009; 364, 2763–2776. https://doi.org/10.1098/rstb.2009.0089
28. Takabayashi T, Mochizuki T, Otani N, Nishiyama K, Ishimatsu S. Anisakiasis presenting to the ED: clinical manifestations, time course, hematologic tests, computed tomographic findings, and treatment. Am J Emerg Med. 2014; 32, 1485–1489. https://doi.org/10.1016/j.ajem.2014.09.010