Acanthocephala from marine fish in the maritime border region of Indonesia and the Philippines

Dhito Dwi Pramardika[1,4], *Fadjar* Satrija[1,2,*], *Sulistiono*[3], *Risa* Tiuria[1,2], and *Arifin* Budiman Nugraha[1,2]

[1]Veterinary Biomedical Sciences Study Program, Graduate School of IPB University, Bogor, Indonesia
[2]Division of Parasitology and Medical Entomology, School of Veterinary Medicine and Biomedical Sciences, IPB University, Bogor, Indonesia
[3]Department of Aquatic Resources Management, Faculty of Fisheries and Marine Sciences, IPB University, Bogor, Indonesia
[4]Department of Health, Nusa Utara State Polytechnic, Sangihe Islands, North Sulawesi, Indonesia

Abstract. A total of 24 fish belonging to seven species were sampled from the waters of the Sangihe Islands in the border area of Indonesia and the Philippines. The fishes were examined to identify the genera of Acanthocephalan phylum parasites that infect marine fish, measure the prevalence and intensity of infection of these parasites, and analyze the correlation of biotic factors with *Acanthocepala* phylum parasite infection in marine fish. Two acanthocephalan genera (*Echinorhynchus* and *Rhadinorhynchus*) were found to infect the marine fish *Aprion virescens* (1/4) and *Katsuwonus Pelamis* (7/7). *Aprion virescens* had a strong correlation between biotic factors such as total length, fish weight and parasite infection, except for the fish condition factor, which had a weak correlation. In contrast, *Katsuwonus pelamis* fish had a strong correlation with weight and fish condition, whereas total length had a weak correlation with parasite infection. It can be concluded that marine fish at the border of Indonesia and the Philippines are infected with *Echinorhynchus* sp. with mild intensity, while *Rhadinorhynchus* sp. is infected with moderate intensity.

1 Introduction

Acanthocephala, a thorny-headed worm, is a metazoan parasite in fish [1]. The parasite possesses a hooked proboscis that attaches to and penetrates the host intestinal mucosa, causing ulceration. In the worst cases, perforation of the intestinal wall and bleeding occur [2]. *Acanthocephala* species have been reported to potentially infect humans. These potential species were suspected to include *B. turbinella, B. nipponicum, B. capitatum, C. villosum, and C. validum*. The life cycle of this parasite is very complex because it requires intermediate hosts, such as amphipods, where larval development takes place, and Teleost fish as the definitive host, where the maturation of adult worms and the reproduction process

* Corresponding author: fadjar_s@apps.ipb.ac.id

occur. In the definitive host, this parasite attaches to the host's intestinal wall with the help of a proboscis. This can damage the intestinal mucosa of fish [3].

Parasitic infections can significantly affect the health and sustainability of individual fishes. The causes of increased parasitic infections are biotic (size, age, and immune status of the host and environment) and abiotic (temperature stress, sedimentation, chemicals, nutrient imbalance, and ultraviolet radiation) factors, which play significant roles [4]. To the best of our knowledge, *Acanthocephala* infection has never been reported in fish from North Sulawesi Province, especially on Marore Island. The island, located in northern Indonesia and directly adjacent to the Philippines, is an area with high marine biodiversity. The area is home to various fish species that play an important role in the marine ecosystem and the local economy.

The aim of this study was to identify fish species infected with *Acanthocephala* parasites and measure the prevalence and intensity of parasite infections in marine fish from Marore Island. Correlations between the number of parasites and total fish length, total fish weight, and fish conditions as biotic factors were also analyzed.

2 Materials and methods

2.1 Study site

The fish were obtained from seven fishing sites in the sea of Marore Island, North Sulawesi Province, on the border between Indonesia and the Philippines. The seven fishing locations were the first point (4.756152°N, 125.513641°E), second point (4.752263°N, 125.508369°E), third point (4.753288°N, 125.507835°E), fourth point (4. 753852°N, 125.506386°E), fifth point (4.753453°N, 125.505264°E), sixth point (4.752303°N, 125.503937°E) and seventh point (4.754039°N, 125.508705°E) (Figure 1). This study was conducted between January and May 2024.

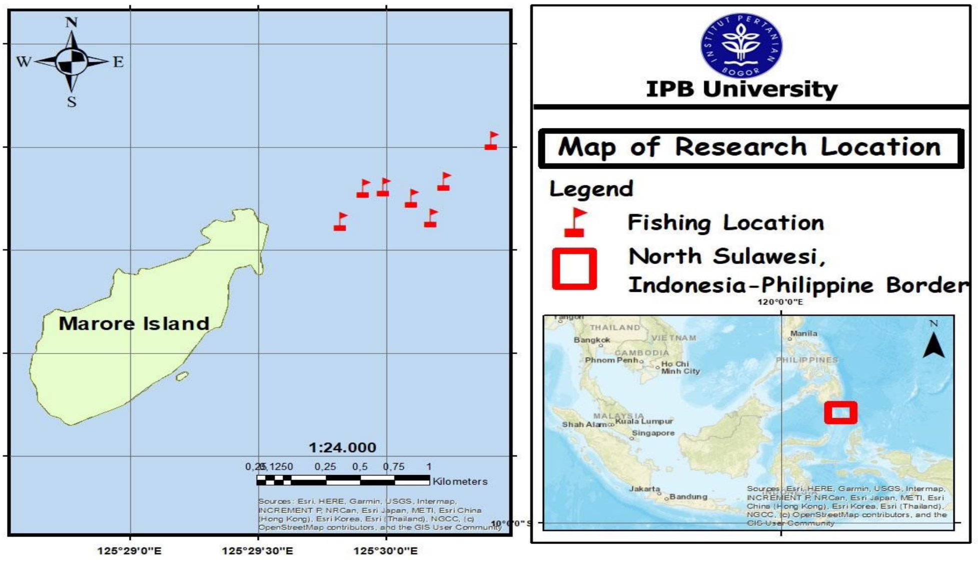

Fig. 1. Research location

2.2 Research Procedure

A total of 24 fish were collected from seven locations. Fish were weighed (g), measured in terms of length (cm), and photographed. Fish samples were stored in a cool box and brought to the laboratory for morphological identification of fish species.

Acanthocephala parasites were collected by incision on the ventral part starting from the cloaca anterior to the operculum to retrieve the digestive organs. The parasites were then washed using NaCl to remove dirt. Morphological identification using an Olympus CX33 microscope with an Indomicro digital camera was performed using Image J. Parasites were stored in 70 % alcohol.

2.3 Data analysis

The prevalence, intensity, and abundance of parasite were determined using the formula of Margolis et al. (1982) [5] as follows:

$$\text{Prevalence} = \frac{NI}{NE} x\ 100\% \text{ ....(1)}$$

$$\text{Intensity} = \frac{TP}{NI} x\ 100\% \text{ ....(2)}$$

$$\text{Abundance} = \frac{TP}{NE} x\ 100\% \text{ ....(3)}$$

Information:
NI = number of hosts infected with parasites
NE = total number of hosts examined
TP = total number of individuals of a particular parasite species in the host sample

The prevalence of infection was categorized as: always (99 – 100 %), almost always (90-98 %), usually (70-89 %), often (50-69 %), commonly (30-49 %, frequently (10-29 %), sometimes (1-9 %), rarely (0.1-1 %), very rarely (0.01-<0.1 %), and almost never (<0.01 %). The infection intensity categories were very mild (<1), mild (1-5), moderate (6-50), severe (51-100), very severe (>100), and superinfection (>1,000) [6].

Correlation analysis between the number of parasites and total fish length, total fish weight, and fish condition as biotic factors was performed using the correlation test. The fish condition value was calculated using the following formula [7]:

$$\text{K} = \frac{W}{aL^b} x\ 100\% \text{........................(4)}$$

Information:
W = fish weight (g)
L = fish length (mm)
a = Constanta
b = length-weight relationship pattern estimator

The condition factor (K) was a condition that states the maturity of the fish and is calculated; in conducting the calculation, the length-weight relationship is analyzed first. K values ranging from to 3-4 indicate that the fish body was slightly flattened. Meanwhile, K values ranging from to 1-3 indicate that the fish body is less flattened. The relationship between length and weight is allometric; it is positive allometric if b is greater than 3 (weight increases faster than length), negative allometric if b is less than 3 (length increases faster than weight increases, and isometric if b is equal to 3 (length growth equals weight) [7].

Spearman's rho rank test was used for the correlation tests. The correlation coefficient (r) describes the level of closeness of the relationship between each variable, which is strong (0.68-1), moderate (0.36-0.67), or weak (>0.36) [8].

3 Results and discussion

Seven fish species were identified in the waters of the Marore Island (Table 1). The seven types of fish are Kerapuh (*Variola louti*), Layang (*Decapterus macrosoma*), Kwee (*Ferdauia orthogrammus*), Kurisi Hijau (*Aprion virescens*), Tuna Gigi Anjing (*Gymnosarda unicolor*), and Cakalang (*Katsuwonus pelamis*). The most common fish species were *K. pelamis*, followed by *D. macrosoma* and *A. virescens*. *A. virescens* was the heaviest (3,006.3 g) and longest (66.8 cm) fish found in this study (Table 1).

Table 1. *Prevalence and infection intensity of Acanthocephala in fish from Marore Island*

Host	AL (cm)	AW (g)	NE	NI	Parasites		P (%)	I	b	ACF
					Rh	Ec				
Variola louti	28 ± 2.83	300 ± 65.05	2	-	-	-	-	-	-6.11	1.54
Decapterus macrosoma	29.4 ± 2.14	253.8 ± 53.85	4	-	-	-	-	-	2.67	1.0
Ferdauia orthogrammus	46.5 ± 3.54	1459.5 ± 412.24	2	-	-	-	-	-	0.76	3.43
Aprion virescens	66.8 ± 4.5	3006.3 ± 374.25	4	1	-	5	25	5	1.82	0.99
Gymnosarda unicolor	58.7 ± 0.58	2354.3 ± 55.16	3	-	-	-	-	-	-2.35	0.98
Elegatis bipinnulata	55 ± 26.87	1927 ± 1026.72	2	-	-	-	-	-	1.05	1.64
Katsuwonus pelamis	49.9 ± 1.57	2119.6 ± 270.52	7	7	151	-	100	21.6	2.23	0.98
Total			**24**	**8**	**151**	**5**	**33.3**			

Noted: AL = Average Length (mean±SD), AW = Average Weight (mean±SD), NE = Number Examined, NI = Number Infected, ACF = Average Condition Factor, Rh= *Rhadinorhynchus* sp., Ec = *Echinorhynchus* sp., P = Prevalence, I = Intensity, b = length-weight relationship pattern estimator

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Fig. 2. Fish species infected with *Acanthocephala* parasites, 1 = *Aprion virescens*, 2 = *Katsuwonus pelamis*.

Two of the seven fish species were infected with *Acanthocephala*. Two infected fish species were *A. virescens* with *Echinorhynchus* sp. infection in five fish, with a prevalence of 25 %, and *K. pelamis* with *Rhadinorhynchus* sp. infection in 151 fish, with a prevalence of 100 % (Table 1, Figures 2, 3, and 4). *Echinorhynchus* sp. and *Rhadinorhynchus* sp. parasites were mostly found in fish guts. The intensity of *Aprion virescens* infection was categorized as mild (5), whereas in *K. pelamis*, it was moderate (21.6) (Table 1).

This study is the first on *Acanthocephala* parasites in marine fishes at the border between Indonesia and the Philippines, providing new insights into the impact of parasites on local fish species. With two species, *K. pelamis* and *A. virescens*, identified as infected, this finding is important given that these fish are the main marine products of Sangihe Regency. *K. pelami*s production totaled 967 thousand kg and *A. virescens* 6 thousand kg, confirming their significant economic and ecological value to the region [9, 10, 11].

Acanthocephala infections from the study results were caused by *Echinorhynchus* sp. and *Rhadinorhynchus* sp. This parasite is not zoonotic. However, there have been many reports of this infection in fishes. Some reports of these parasitic infections occurred in North Sumatra, with *Rhadinorhynchus* sp. and *Echinorhynchus* sp. infections found in 100% of *K. pelamis* intestines [17]. *Rhadinorhynchus* sp. infection was also reported in *Chanos chanos* in Banda Aceh City at 10 %, with microhabitats at 33 % in the intestine and 7 % in the esophagus [18]. *Rhadinorhynchus* sp. infection was also reported to be 13.3% in *Euthynnus affinis* from the Sunda Strait [19].

Several reports of *Rhadinorhynchus* sp. and *Echinorhynchus* sp. infections have also been reported in other countries, such as *Rhadinorhynchus* sp. infection in *K. pelamis* in Batangas Sea Philippines [20]. *Echinorhynchus* sp. infection has also been widely reported, such as in India in *Thunnus albacares* at 52.67 % with an intensity of 12.46 [21]. Japan reported *Liparis* sp. as the first host of *E. hexagrammi* infection in the gut [22]. To date, no cases of *Echinorhynchus* sp. infection have been reported in *A. virescens*. This is a new report on *Echinorhynchus* sp. infection in *A. virescens*. Studies have documented parasitic infestations in *A. virescens*, with *Monogenea* and *Digenea* found to affect the gills and abdominal regions [23]. *Digenea* species reported to infect are *Pleorchis uku*, *Metadena rotivarijera*, *Maculifer aprionis*, and *Brachyenteron microvaei* [24].

Rhadinorhynchus sp. has been widely reported to infect *K. pelamis*. This further strengthens the results of our study, namely, the suspicion that *K. pelamis* is a host specific to *Rhadinorhynchus* sp., and its microhabitat is in the intestine. Based on their life cycle, they live as parasites in crustaceans during their larval stage. In contrast, in the adult stage, they inhabit the invertebrate digestive tract, especially the digestive organs of fish [25]. Digestive organs become microhabitats for *Acanthocephala* worms by providing food that is readily metabolized by their bodies [26].

Table 2. Morphometric and microhabitat of parasite *Acanthocephala* (mean ± SD)

Parasites	BL	PL	LSP	MH
Echinorhynchus sp.	9077.17 ± 19.83	462.19 ± 4.82	49.99 ± 5.78	Intestine
Rhadinorhynchus sp.	22379.49 ± 1432.57	1607.35 ± 945.44	57.71 ± 16.81	Intestine

BL = Body Length (μm); PL = Proboscis Length (μm); LSP = Length of the Spines on the Proboscis (μm); MH = Microhabitat

The results of the fish growth analysis of all samples showed negative allometric growth, including *A. virescens* and *K. pelamis*, infected with *Acanthocephala* parasites with b values of 1.82 and 2.23, respectively. This indicates that fish on Marore Island grow their body length faster than their weight gain. Regarding the average condition factor, *A. virescent* (0.99) and *K. pelamis* (0.98) on Marore Island exhibited poor conditions (Table 1). This phenomenon can be caused by *Acanthocephala* parasite infection, which disrupts the metabolic process and health of fish, environmental conditions that may not support optimal growth, and limited food availability, which encourages fish to grow longer without balanced weight gain [21].

Based on the environment and food availability, the marine area of Marore Island has a natural habitat that is favorable for these fish to live and breed. A report states that the sea temperature on Marore Island ranges from to 20-24 °C with a salinity of 34.80-35.20 and has an abundance of phytoplankton from the genus *Isthmia nervosa,* and is rich in zooplankton from the *Crustacea class* (75 %) and *Gastropoda* (25 %) [12]. This is supported by research that states that the habitat ranges from to 20.5-26 °C [13]; and usually consumes smaller fish; they can also consume c*rustaceans* [14]. *Crustaceans* are common intermediate hosts for *Acanthocephala*. Its life cycle begins when the host releases eggs into water. The intermediate host then eats the eggs and hatches in the intestinal lumen into *acanthors*, penetrates the intestinal wall, and attaches to the serosa within 2-4 hours. *Acanthors* were found to be free in the hemocoel and metamorphosed to the Acanthella stage for up to 12 days in the intermediate host. *Acanthella juveniles* become infective to the definitive host on days 13-14 [15]. Some experimental studies have shown that temperature is an important factor in the development of *Acanthocephala* species. Infective development occurs for 30-32 days at 20-25°C, whereas the infective stage of *Echinorhinchus truttae* takes 82 days at 10-20°C. In general, *Echinorhinchus* sp. became infective in 13 days at 21-23°C [16]. The possibility of negative allometric fish body height is high prevalence and intensity of *Acanthocephala* infections in the host can adversely affect fish health. This parasite can cause intestinal obstruction and trigger an inflammatory response, which can lead to colitis. Emaciation is a common symptom in fish infected with this parasite [30].

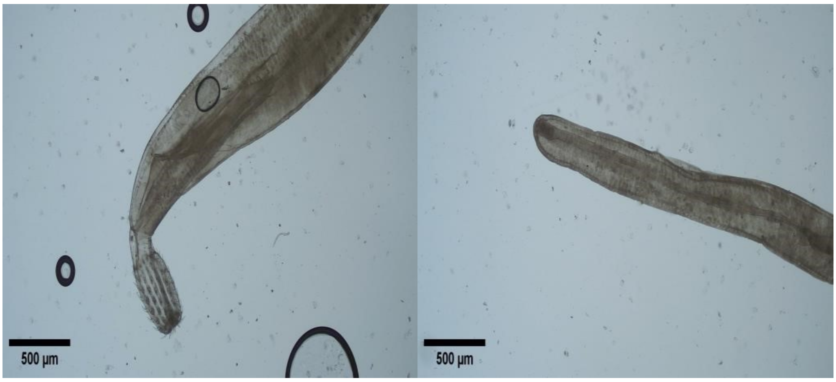

Fig. 3. *Echinorhynchus* sp. infection in *Aprion virescens* fish, 1 = Anterior with 40x magnification, P = Proboscis, 2 = Posterior with 40x magnification, F = Female sex.

The *Echinorhynchus* sp. found on *A. virescens* was identified as a female. It was visible on the posterior part (Figure 3), then the average length of this parasite reached 9077.17 µm with a proboscis length of 462.19 µm and the average length of spines in the proboscis of 49.99 µm (Table 2). At the same time, *Rhadinorhynchus* sp. on *K. pelamis* was identified as female (figure 4), with an average length of 22379.49 µm with a proboscis length of 1607.35 µm and an average length of spines in the proboscis of 57.71 µm (Table 2).

The morphometric results in this study were not significantly different from those of other studies. The length of the parasite depends on the host and intensity of the parasite infection [27]. Morphometric characteristics of *Rhadinorhynchus* sp. in *Auxis thazard* are mentioned with a body length of 4.75 - 11.25 mm for males, while females are 7.80 - 26.25 mm. The proboscis was then separated by an aspinose part of the anterior part of the parasite. The posterior part of the female was longer than the male [28]. *Echinorhynchus* sp. in *Hippoglossus stenolepis* has a body length of 4.5 - 23 mm, whereas in females, it has a length of 10.5 - 82.90 mm. In addition, mature eggs are present in the body cavities of the female worms. The posterior part of the body consists of the uterus and uterine bell [29].

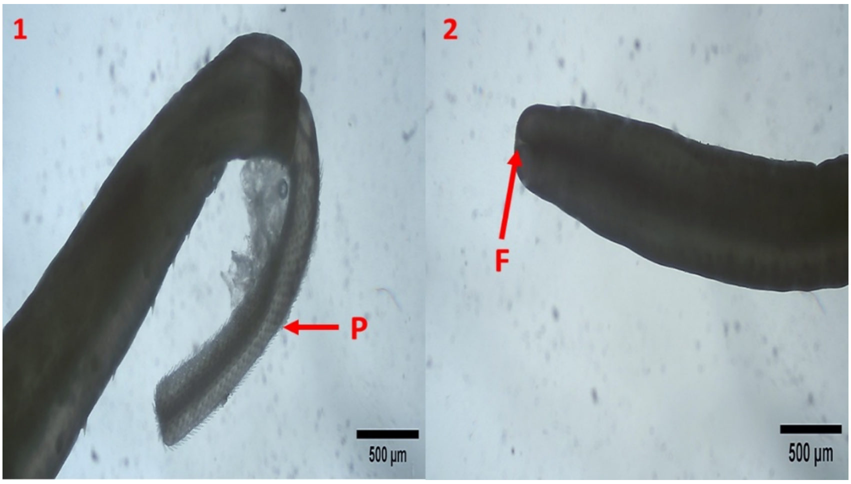

Fig. 4. *Rhadinorhynchus* sp. infection in *Katsuwonus pelamis* fish, 1 = Anterior with 40x magnification, P = Proboscis, 2 = Posterior with 40x magnification, F = Female sex

Table 3. Spearman's rho correlation test between the number of parasites and biotic factors

Host	Biotic factor	R	Correlation
Aprion virescens	Total length	0.816	Strong
	Weight	0.775	Strong
	Condition factor	-0.258	Weak
Katsuwonus pelamis	Total length	-0.259	Weak
	Weight	-0.857*	Strong
	Condition factor	-0.679	Strong

* significant (p-value < 0.05)

The negative correlation value of the fish condition factor with the number of parasites (Table 3) indicates a tendency for high parasite infection in fish with poor condition factors of the fish itself. Biotic factors, such as total length (0.816) and weight (0.775), in *A. virescens* were strongly correlated with parasite infection, whereas the fish condition factor (-0.258) was a weakly correlated. *K. pelamis*, based on the correlation value, weight (-0.857), and fish condition factor (-0.786), had a strong correlation with parasite infection, whereas the total length (0.259) had a weak correlation value. The biotic factors were strongly correlated. This is in line with research that states that the size and weight of the host body greatly affects parasitic infection [31]. In addition, the condition of fish obtained by calculating body length-mass is related to parasite infection, as described in a study stating that the condition factor of fish not infected with parasites is higher than that of those not infected [32]. The condition factor describes the growth of fish. If the condition of the fish is low, then the fish is susceptible to parasite infection, which results in the fish not experiencing isometric growth [31].

4 Conclusion

In this study, marine fish species were infected with *Acanthocephala* parasites at the border of Indonesia and the Philippines and two genera of parasites infecting these fish, *Echinorhynchus* sp. and *Rhadinorhynchus* sp. The prevalence of infection was significant, with a clear correlation between biotic factors, such as length, weight, and condition of the fish, and the level of infection. These findings provide important insights into the effect of *Acanthocephala* parasites on marine fish health. The two infected fish species belong to the commercial fish category; therefore, further research on infection mechanisms and the environment is needed for more effective management strategies and fishery sustainability.

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