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MONOGENEAN INFECTIONS IN RABBITFISH (*Siganus* spp.) FROM THE CAT BA ARCHIPELAGO VIETNAM

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ABSTRACT

A total of 133 white-spotted spinefoot (*Siganus canaliculatus*) and 80 mottled spinefoot (*Siganus fuscescens*) were examined between 2009 to 2025 to determine the prevalence, intensity, and species composition of monogenean parasites. Monogeneans were isolated, mounted by gelatin-glycerin, and stained with either carmin alum or trichome gomori for morphological identification. The infection rate in both fish species was high (> 75%), varying by sampling period (year). The mean intensity of infection in *S. canaliculatus* was 12.3 parasites per host (range: 1–100), which was higher than in *S. fuscescens* at 8.0 (range: 1–36). Six species of monogeneans were identified: *Glyphidohaptor sigani* Kritsky, Galli & Yang, 2007; *Glyphidohaptor* sp.1; *Glyphidohaptor* sp.2; *Tetrancistrum indicum* (Paperna, 1972) Kritsky, Galli & Yang, 2007; *Tetrancistrum sigani* Goto & Kikuchi, 1917 and *Polylabris mamaevi* Ogawa & Egusa, 1980. Five species were found in *S. canaliculatus*, five in *S. fuscescens*, and four species were shared between both hosts. These findings represent the first confirmed records of monogeneans in Vietnamese rabbitfish.

Keywords: Monogenea, parasite, *Siganus canaliculatus*, *Siganus fuscescens*, rabbitfish, Cat Ba, Vietnam.

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INTRODUCTION

The family Siganidae Richardson, 1837, currently includes 29 valid rabbitfish species, mainly distributed throughout the Indian Ocean - Western Pacific (Froese & Pauly, 2025). Rabbitfish typically appear in pairs or flock around coastal and coral reefs, generally inhabiting depths of 15 meters and above. While only a limited number of rabbitfishs are used for human consumption, others are kept in aquaria due to their vibrant coloration. These fish species are often raised with other economically important fish species to take advantage of food sources.

Rabbitfish enhabit diverse habitats and display a diversity of feeding behaviors, which makes them highly susceptible to parasites. However, only eight rabbitfish species have been reported as hosts to monogenean (Kritsky et al., 2007a, b; Al Jufaili & Palm, 2017; Al Jufaili et al., 2020): *Siganus canaliculatus* (Park, 1797), *Siganus corallinus* (Valenciennes, 1835), *Siganus doliatus* Guérin-Méneville, 1829, *Siganus fuscescens* (Houttuyn, 1782), *Siganus lineatus* (Valenciennes, 1835), *Siganus luridus* (Rüppell, 1829), *Siganus punctatus* (Schneider & Forster, 1801) and *Siganus rivulatus* Forsskål & Niebuhr, 1775. A total of 23 monogenean species have been identified in rabbitfish, including genera: *Glyphidohaptor* Kritsky, Galli & Yang, 2007; *Pseudohaliotrema* Yamaguti, 1953 and *Tetrancistrum* Goto & Kikuchi, 1917.

In Vietnam, rabbitfish are also an important species in aquaculture and are used as ornamental fish. Some large-sized, delicious-fleshed species are raised semi-intensively, such as *Siganus guttatus*, *S. canaliculatus*, and *S. fuscescens*, which are often raised as ornamental fish. Consequently, their parasitic fauna has attracted the attention of many Vietnamese researchers (Amin & Nguyen, 2011; Nguyen Ty et al., 2018; Nguyen & Hoang, 2018; Hoang et al., 2017). For example, Nguyen et al. (2018) identified six parasite species (including 3 protozoa, 1 metacercariae, and 2 parasitic copepods) in 300 mottled spinefoots (*S. fuscescens*) farmed from Quang Dien (Hue City). However, no monogeneans were detected (Nguyen et al., 2018).

The Cat Ba Archipelago and Ha Long Bay area, recognized as a UNESCO World Biosphere Reserve in 2004, encompasses diverse marine ecosystems, such as coral reefs, mangroves, and seagrass, to name just a few. These rich environments not only support diverse marine animals but also foster the development and spread of various fish parasites.

MATERIALS AND METHODS

Materials

Rabbitfish are widely distributed along Vietnam's coastal regions, farmed and harvested year-round, which facilitated sample collection. In the Cat Ba archipelago, rabbitfish are caught by bottom trawling and traps at night and transported to the fishing port and fish market in the early morning. After a significant sampling effort (years), we have only encountered two species of rabbitfish: white-spotted spinefoot (*S. canaliculatus*) and mottled spinefoot (*S. fuscescens*) (Table 1).

Rabbitfish were bought from local fishers using bottom trawls or traps in the waters surrounding Cat Ba Island. The fish were preserved in ice-containers and immediately transported to the Parasitology Laboratory, Institute of Biology, Vietnam Academy of Science and Technology for measurement, photographed, and identified to species based on classification references (FishBase; Nguyen Khac Huong, 2022).

Methods

Isolation, processing, and preservation of monogeneans: Fish specimens were examined for parasites (Buchmann, 2007). Monogenean samples were isolated with the aid of a dissecting microscope and fixed in 4% formalin or 70% ethanol.

Glycerin-gelatin mounting: This technique was used to observe the haptoral hooks, anchors, and copulatory structures of small monogeneans. Parasites were arranged on a slide and air-dried for 2–5 minutes. One drop of hot glycerin-gelatin was applied and a cover slide was added; slides were labelled and left to dry for 2–3 hours before storage.

Stained samples: Specimens were fixed in 4% formalin or 70% ethanol for at least two weeks. The samples were stained by alum carmine or Gomori's trichrome. Alum carmine enhances internal organ visibility, while Gomori's stain is more effective at highlighting chitinized structures.

Figure 1. White-spotted spinefoot (*Siganus canaliculatus*)

Figure 2. Mottled spinefoot (*Siganus fuscescens*)

Carmine alum stain: Monogeneans were put in the solution for 10–30 minutes. The samples were decolorized in acid HCl-ethanol (70%). Dehydration followed a series of ethanol solutions (70, 80, 90 and 100%), followed by clearing with xylene and mounting in Canada balsam.

Gomori's trichrome stain: Monogeneans were stained for 1–2 minutes, then immediately placed in 70% ethanol to avoid overcoloring. Dehydration, clearing, and mounting were performed as in the alumi carmine method.

Data processing

Data were recorded in Excel (Microsoft Office 2010) and using Stata IC12.1 (StataCorp LP). Monogenean prevalence was analyzed using logistic regression models, with host species and sampling collection period as predictors. Infection intensity was analyzed using a generalized negative binomial regression, with monogenean counts log-transformed [log(x+1)] to normalize the distribution. Statistical significance was considered at $P < 0.05$.

RESULTS

Monogenean infection status in rabbitfish

A total of 213 specimens from two rabbitfish species were collected in March 2009, 2017, 2019, 2023, 2025, and in October 2024 (Table 1).

Table 1. Number of rabbitfish, representing two species, collected in Cat Ba Island

Name of fishes	Collection time						Total
	March, 2009	March, 2017	March, 2019	March, 2023	October, 2024	March, 2025	
White-spotted spinefoot (*Siganus canaliculatus*)	15	15	15	36	25	27	133
Mottled spinefoot (*Siganus fuscescens*)	13	10	10	17	20	10	80
Total	28	25	25	53	45	37	213

A high prevalence of monogenean infection was observed, with 83.5% in *S. canaliculatus* and 77.5% in *S. fuscescens* (Table 2). Statistical analysis showed no significant differences in infection rates between the two species or among sampling periods across years ($P = 0.129$ and $P = 0.514$, respectively). Previous studies on *S. fuscescens* and *S. guttatus* in Vietnam reported no monogenean infections (Nguyen & Hoang, 2018; Nguyen et al., 2018). Therefore, this study provides the first data on monogenean infections in rabbitfish in Vietnam.

Table 2. Monogenean infection rates in rabbitfish species from Cat Ba Island

Collection Time	*Siganus canaliculatus*			*Siganus fuscescens*		
	∑ fish examined	∑ fish infected	Rate (%)	∑ fish examined	∑ fish infected	Rate (%)
March, 2009	15	14	93.3	13	10	76.9
March, 2017	15	12	80.0	10	8	80.0
March, 2019	15	12	80.0	10	8	80.0
March, 2023	36	27	75.0	17	13	76.5
October, 2024	25	20	80.0	20	17	85.0
March, 2025	27	26	96.3	10	6	60.0
Total	**133**	**111**	**83.5**	**80**	**62**	**77.5**

Statistical analysis of monogenean infection intensity revealed significant differences between the two fish species ($P = 0.05$) and among sampling periods ($P < 0.05$). The mean intensity in *S. canaliculatus* ranged from 8.7−16.7 worms per fish, while in *S. fuscescens* it ranged from 3.2–14.8 (Table 3). No significant differences were found between the two seasons in 2024 ($P > 0.05$).

Table 3. Monogenean intensity in rabbitfish species from Cat Ba Island

Collection time	*Siganus canaliculatus*		*Siganus fuscescens*	
	Intensity	∑ worms	Intensity	∑ worms
March, 2009	10.5 (1–61)	147	3.2 (1–12)	32
March, 2017	14.8 (5–27)	177	5.8 (1–23)	46
March, 2019	10,4 (1–8)	125	3.8 (1–8)	30
March, 2023	8.7 (1–24)	234	14.8 (1–31)	193
October, 2024	12.3 (1–32)	246	8.7 (1–25)	148
March, 2025	16.7 (1–100)	435	8.0 (1–36)	48
Total	12.3 (1–100)	1364	8.0 (1–36)	497

Note: Intensity - Number of monogeneans/infected fish.

Monogenean species composition in hosts

Based on the examination of gelatin-glycerin mounted and stained specimens, seven monogenean species were identified. Five species (Monopisthocotylea) belonging to two genera of Ancyrocephalidae Bychowsky, 1937: *Glyphidohaptor sigani* Kritsky, Galli & Yang, 2007; *Glyphidohaptor* sp.1; *Glyphidohaptor* sp.2; *Tetrancistrum indicum* (Paperna, 1972) Kritsky, Galli & Yang, 2007; *Tetrancistrum sigani* Goto & Kikuchi, 1917 and the only species, *Polylabris mamaevi* Ogawa & Egusa, 1980 (Polyopisthocotylea) was found in both rabbitfish species *Piluabris mamaevi* is easily identified by its large body size and clamp structure.

Monogenean specimens of *Glyphidohaptor* are small in size and very difficult to identify to species. Therefore, gelatin-glycerin-mounted slides are necessary to improve the visibility of key morphological features, such as the male copulatory structures, bars, and anchors. The two species of *Tetrancistrum* are similar in size but can be clearly distinguished by observing their copulatory complexes under a microscope (400x magnification).

The infection rate of monogeneans (2−3 species) was rather high, ranging from 30.63% to 36.04% in *S. canaliculatus*, and 32.26% to 35.48% in *S. fuscescens* (Table 4). Moreover, this study shows that nine individuals of the white-spotted spinefoot were infected with up to five monogenean species (8.1%) out of 111 examined fish, while five species of monogeneans (6.45%) simultaneously parasitised 4 individuals out of 63 infected

mottled spinefoot individuals. Interestingly, we observed that very closely related species co-inhabited the same host with minimal interspecific competition. Furthermore, the differences in body size may contribute to reduced competition within the gill zones. Observations made under a dissecting microscope during the survey indicated that monogeneans were concentrated on the outer gill filaments.

Table 4. Infected rate of monogenean species in rabbitfish

Name of fishes	Number of monogenean species and infected rate (%)				
	1 species	2 species	3 species	4 species	5 species
Siganus canaliculatus	13.51	36.04	30.63	11.71	8.10
Siganus fuscescens	16.13	35.48	32.26	9.68	6.45

DISCUSSION

The prevalence and intensity of monogenean infections in rabbitfish species were very high, largely due to environmental and life conditions that are favorable for the invasion of monogenean species. *S. fuscescens* live in schools within algae and seagrass beds, shallow lagoons, coastal coral reefs, and in large estuaries. Juveniles feed on filamentous algae, adults on foliar algae and seagrass (Devi & Rao, 2003). They are active throughout the day, increasing their exposure to monogeneans. Adult white-spotted spinefoots (*S. canaliculatus*) live in coastal areas, algal reefs, estuaries and in large lagoons with algal-crustal habitats, mainly common on rocky substrates, feeding on algae and seagrass. Unlike *S. fuscescens*, this species is more tolerant of turbid water, occurring in the vicinity of estuaries, especially around seagrass beds. Adults may also venture a few kilometres offshore into deeper water. Juveniles form very large schools in shallow bays and reefs; school size decreases with size, with adults occurring in groups of about 20 individuals (Woodland, 1997). This schooling behavior facilitates high parasite transmission, such as monogeneans, due to increased opportunities for cross-infection. Additionally, migratory habits and wide foraging areas of rabbitfish further increase their exposure to parasites.

The monogenean species identified from two rabbitfish species in this study are very similar, with 4/6 species found on both, except for *Glyphidohaptor* sp.1, which was found only on *S. canaliculatus*, while *Glyphidohaptor* sp.2 was found only on *S. fuscescens*.

Until now, *Glyphidohaptor* Kritsky Galli & Yang, 2007 was reported exclusively to parasitize rabbitfish (Kritsky et al., 2007a). It has been found on a wide range of rabbitfish species, such as these colorful reef-associated species: *S. corallinus*, *S. doliatus*, *S. lineatus* and *S. punctatus* and non-reef, drab-colored species: *S. luridus*, *S. fuscescens* and *S. rivulatus* (Jufaili et al., 2020).

The genus *Tetrancistrum* Goto & Kikuchi, 1917 is currently comprised of 17 known species. Although the morphology of the members of the genus is homogeneous, they are distinguished primarily by their copulatory complexes, and therefore is the primary mechanism for identifying species of the genus (Jufaili & Palm, 2017). Since the original description of *T. sigani* from the gills of *S. fuscescens* (Goto & Kikuchi, 1917), at least seven species have been described at various localities (Young 1967; Paperna 1972; Martens & Moens 1995; Kritsky et al. 2007b).

Another monogenean genus, *Polylabris* Euzet & Cauwet, 1967, is also found on the gills of fishes: *Polylabris sigani* Dillon, Hargis & Harrises, 1983 in *Siganus oramin* (= *S. canaliculatus*) and *P. mamaevi* Ogawa & Egusa, 1980 in *S. stellatus* (Tingbao et al. 2007). *Polylabris bengalensis* Sailaja & Madhavi, 2011 is described from *S. canaliculatus* and *S. javus* (Sailaja & Madhavi, 2011).

The genus *Polylabris* Euzet & Cauwet, 1967 includes 2 species reported from Vietnam: *Polylabris gerres* (Sandars, 1944)

Mamaev & Parukhin, 1976 from silverbiddy fish (*Gerres lucidus, Gerres filamentosus*) and *Polylabris tubicirrus* (Paperna & Kohn, 1964) Mamaev & Parukhin, 1976 from the toothpony fish (*Gazza minuta*) and the pugnose ponyfish (*Leiognathus insidiator*). *P. mamaevi* Ogawa & Egusa, 1980, is recorded for the first time in both fish species in this study.

CONCLUSION

Prevalence and intensity of monogenean infections in rabbitfish from Cat Ba Island were high (> 75%; 8.74 worms/fish). Mean intensity of infection in *S. canaliculatus* was 12.3 (range of 1–100), and higher than in *S. fuscescens* at 8.0 (range of 1–36). Six monogenean species were identified: *G. sigani* Kritsky, Galli & Yang, 2007; *Glyphidohaptor* sp.1; *Glyphidohaptor* sp.2; *T. indicum* (Paperna, 1972) Kritsky, Galli & Yang, 2007; *T. sigani* Goto & Kikuchi, 1917; and *P. mamaevi* Ogawa & Egusa, 1980. These results are the first confirmed records of monogeneans in Vietnamese rabbitfish. Further morphological and molecular analyses are needed to determine if the undescribed species are new to science.

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