Ectoparasite attack on star pomfret (*Trachinotus blochii*) at Batam Mariculture Center, Indonesia

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Abstract. Star pomfret (*Trachinotus blochii*) has great market potential to be developed, both for domestic and international market demands. Parasitic infections in these fish can trigger or cause a disease and even death. Ectoparasites are parasites that inhabit the external organs of the host. This study aimed to identify ectoparasites in star pomfret cultured at the Batam Mariculture Center (BMC) Batam, Indonesia. It was carried out from January to April 2022. Samples of fish that were indicated to be sick were randomly selected once a week. A total of 25 fish samples were taken from 7 rearing containers (floating net cage, nursery tank and fish tank). Observations of fish morphology and clinical symptoms included organ integrity, body color, mucus on the skin surface, gills, operculum, eyes, mouth and fins. The identification of ectoparasites was carried out using the wet mount method. Samples were observed under a microscope and the characteristics obtained were matched with a parasite identification book. In this study, identified ectoparasites in star pomfret were *Benedenia* sp. (Phylum: Platyhelminthes) on the skin, *Amyloodinium* sp. (Phylum: Myzozoa) on the gills, *Trichodina* sp. (Phylum: Ciliophora) on the gills, and *Diplectanum* sp. (Phylum: Platyhelminthes) also on the gills.
Key Words: *Amyloodinium* sp., *Benedenia* sp., *Diplectanum* sp., parasite, snubnose pompano, *Trichodina* sp.

Introduction. One of the fish commodities that are cultivated in Indonesia is star pomfret or snubnose pompano (*Trachinotus blochii*) which has great potential and a promising market, both at home and abroad. Although relatively new, star pomfret has been able to attract the attention of cultivators. This is because the star pomfret has fast growth, resistance to disease, easy maintenance, and high market demand (Haryanto et al 2021).

Diseases in fish are one of the obstacles that cause failure in aquaculture business (Hidayatullah et al 2020). In general, diseases in marine fish farming can be divided into 2 groups, namely infectious and non-infectious diseases. Infectious diseases are caused by pathogenic organisms and are capable of spreading through the movement of an infected host. This group of diseases can be divided into 4 subgroups, namely parasitic, bacterial, viral, and mycotic diseases. While non-infectious diseases are generally caused by environmental conditions, nutritional deficiencies, genetics, poor management of cultivation activities, and contamination from toxic compounds (Dadar et al 2017; Attademo et al 2020).

The impact caused by parasites on fish is actually no less important than bacteria and viruses. If the intensity is large, the fish that are kept will get sick easily and their growth will be stunted. The scars on the body, namely the place where the parasite attaches will cause lesions so that other disease agents in the form of viruses and bacteria will easily attack. In addition, when the fish's immune system is disturbed, it will affect the fish's appetite so that the growth rate is inhibited (Kabata 1985; Luo et al 2008).

Based on the place of landing, they are divided into two, namely endoparasites and ectoparasites. Endoparasites are types of parasites that live in the body of the host such as the intestines, kidneys, and liver. While ectoparasites are parasites that live and

attach to the outer body of the fish such as gills, skin, and fins. Parasites have been widely known and the losses they cause are also very diverse. Ectoparasites, a group of organisms living on the surface of the skin, may cause irritation and inflammation or various forms of allergic reactions and other discomfort and anxiety to the host (Johnson et al 2004; Sitjà-Bobadilla et al 2021).

The survival rate of star pomfret at the nursery stage is still quite low due to disease attacks that interfere with production (Kalidas et al 2012; Alejos Jr. & Serrano 2018; Putra & Raza'i 2020). Hakim et al (2019) successfully isolated parasites from star pomfret cultivated in neighbour island, Tanjung Pinang waters, Indonesia, which were *Derogenes varicus*, *Lecithochirium* sp., *Gorgonorhynchus* sp., *Ornithonyssus* sp. and *Camalanus* sp.. Palma et al (2022) reported the parasites *Lepeophtheirus spinifer* and *Neobenedenia* sp. coinfected with star pomfret in floating net cages in the Philippines. Adult male (70-100%) and female (90-100%) *L. spinifer* have a high prevalence in both winter and summer. While coinfection by *Neobenedenia* sp. occurs throughout the year without significant changes in population intensity for each season.

The same thing was reported by Pakingking Jr. et al (2018), where symptoms of exophthalmia were seen among star pompret brooders cultured with the Kaligid parasite (*L. spinifer*) in Philippines. After sequencing, and based on the results of diagnostic investigations and experimental infection, *Vibrio harveyi* may have had a role in the reported cases of exophthalmia, and this was initiated by *L. spinifer* infection. Nam et al (2020) reported the presence of skin worms on star pompret and identified them to the species level by morphology and molecular analysis. Clinical symptoms shown include dyspnea, anorexia and light bleeding on the skin. All fish samples were infected with worms on the skin, gills and eyes, covered with excessive mucus. This study aimed to identify ectoparasites in star pomfret (*Trachinotus blochii*) which were cultured at the Batam Mariculture Center (BMC), Batam, Province of Riau Islands, Indonesia.

Material and Method

Time and places. The research was carried out from January to April 2022 in BMC, Batam, Province of Riau Islands, Indonesia.

Tools and materials. The tools used were analytical scales, object glass, cover glass, bucket, tissue, microscope (Olympus OP73 binocular microscope), dropper, tray, surgical scissors, tissue, drain, petri dish, and ruler. Meanwhile, the materials used were star pomfret fish, sized 3.5-43.5 cm total length, hatched and cultivated in BMC, Batam. Physiological solution (0.9% NaCl) was used as a medium for observing parasites.

Research methodology. Large pompret starfish were reared in floating net cage (FNC) (size 4 x 4 x 4 m). Density varied between 500 and 2000 fish cage^{-1} and fed with fresh fish and trash fish as much as about 5% of the fish body weight. There were 50 FNCs available at BMC, Batam. Brood stock was reared in large concrete brood tanks (255 m^3) with a stocking density of 20-50 fish per tank and fed with special pellets for star pompret brood stock. Several square concrete tubs (10 m^3) and fiberglass main tanks (15 m^3) were also used for brood stock rearing in preparation for spawning. The rearing activities of the larvae used a fiberglass tub or nursery tank (1 m^3) and a fish tank (3 m^3) in a hatchery. Larvae were fed with special pellets for larvae, 3 times a day as much as 2-3% of fish body weight. Larval stocking density ranges from 100 to 1500 fish per fish tanks. Several nursery tanks and fish tanks were available and used for the activities.

Fish samples were taken randomly from FNC, nursery tanks and fish tanks that indicated have been infected by the parasites. The suspected tanks containing infected fish were determined based on the experience of researchers from BMC. The sampling was carried out once a week. The characteristics and morphology as well as clinical symptoms of the sampled pomfret population were as shown in Table 1. A total of 25 fish samples were taken from 7 rearing containers, namely FNC no. 27 (4 fish), FNC no. 5A (2 fish), FNC no. 43 (2 fish), FNC no. 44 (4 fish), fish tank no. 5 (1 fish), nursery tank 1 (3 fish), and nursery tank 3 (9 fish). The fish samples were observed for morphological and

clinical symptoms, both for sick and dead fish and includes completeness of organs, body color, gills, operculum, eyes, mouth and fins, including the condition of body mucus, gills and eyes. Identification of ectoparasites in star pomfret was carried out using the wet mount method (Khanna et al 2014). Samples of parasites were taken from the gills, fins, mucus on the body surface. Furthermore, the sample was placed on a glass object and then dripped with physiological NaCl solution and then covered with a cover glass. Then the sample was observed under a microscope with a magnification of 40X and 100X and observed carefully for classification. The identification of ectoparasites was based on the books of parasite of fish and other aquatic biota (Kabata 1985; Sitjà-Bobadilla et al 2021). The data obtained were presented in tabular form which was then analyzed descriptively based on the related literature.

Table 1

Morphology and clinical symptoms of sampled star pomfret

No.	*Fish container*	*Total sample*	*Fish clinical symptoms and population behavior*
1	FNC no. 27	4	Many fish died in the cages and there was a lot of mucus on the surface of the sample fish skin.
2	FNC no. 5A	2	Many of the fish look weak, sluggish and often rise to the surface of the water.
3	FNC no. 43	2	Many fish died, the fish seemed to swim weakly and many of them were isolated from their groups in the FNC.
4	FNC no. 44	4	The fish often rised to the surface, rub their bodies into the FNC nets and appear to be swimming slowly.
5	Fish tank no. 5	1	Some fish look convulsive, on the surface of the body there is a lot of mucus.
6	Nursery tank 1	3	Seen some dead fish and some fish seem to swim slowly.
7	Nursery tank 3	9	Seen some dead fish and some fish seem to swim slowly.

FNC: floating net cage.

Results. Based on the observation, parasites were found in the gills (3 samples) and skin (8 samples). Meanwhile, in the mucus samples, no parasites were found. These parasites generally attack fish with larger sizes and are not present in seed size (Table 2). Most of the attacks occurred on fish kept in FNC and only 1 case was found in fish kept in fish tanks. The parasite found on the surface of the fish skin was *Benedenia* sp. (Figure 1) which infected 8 fish. The parasites found on the fish gills were *Amyloodinium* sp., *Trichodina* sp., and *Diplectanum* sp. (Figure 2).

Table 2

Presence and identification of parasites in star pomfret at BMC, Batam

No.	*Sample code*	*Total length of fish (cm)*	*Origin of sample*	*Organ*	*Result*
1	I10/1/22	19.0	FNC 27	Gill	Negative
				Mucus	Negative
				Skin	Negative
2	I11/1/22	18.0	FNC 27	Gill	*Trichodina* sp.
				Mucus	Negative
				Skin	Negative
3	I12/1/22	18.5	FNC 27	Gill	Negative
				Mucus	Negative
				Skin	Negative
4	I13/1/22	18.0	FNC 27	Gill	Negative
				Mucus	Negative
				Skin	Negative
5	I23/1/22	19.0	FNC 5A	Gill	Negative
				Mucus	Negative
				Skin	*Benedenia* sp.
6	I24/1/22	26.0	FNC 5A	Gill	Negative
				Mucus	Negative
				Skin	*Benedenia* sp.

7	I30/1/22	17.5	FNC 43	Gill	Negative
				Mucus	Negative
				Skin	*Benedenia* sp.
8	I31/1/22	17.0	FNC 43	Gill	Negative
				Mucus	Negative
				Skin	*Benedenia* sp.
9	I40/1/22	20.0	FNC 44	Gill	Negative
				Mucus	Negative
				Skin	*Benedenia* sp.
10	I41/1/22	20.5	FNC 44	Gill	Negative
				Mucus	Negative
				Skin	*Benedenia* sp.
11	I47/1/22	18.2	FNC 44	Gill	Negative
				Mucus	Negative
				Skin	*Benedenia* sp.
12	I48/1/22	16.0	FNC 44	Gill	*Amiloodinium* sp., *Diplectanum* sp.
				Mucus	Negative
				Skin	*Benedenia* sp.
13	I20/1/22	43.5	Fish tank 5	Gill	*Amyloodinium* sp.
				Mucus	Negative
				Skin	Negative
14	I25/1/22	3.5	Nursery tank 1	Gill	Negative
				Mucus	Negative
				Skin	Negative
15	I26/1/22	3.5	Nursery tank 1	Gill	Negative
				Mucus	Negative
				Skin	Negative
16	I27/1/22	4.0	Nursery tank 1	Gill	Negative
				Mucus	Negative
				Skin	Negative
17	I27/1/22	4.0	Nursery tank 3	Gill	Negative
				Mucus	Negative
				Skin	Negative
18	I28/1/22	3.2	Nursery tank 3	Gill	Negative
				Mucus	Negative
				Skin	Negative
19	I36/1/22	4.5	Nursery tank 3	Gill	Negative
				Mucus	Negative
				Skin	Negative
20	I37/1/22	3.9	Nursery tank 3	Gill	Negative
				Mucus	Negative
				Skin	Negative
21	I38/1/22	4.5	Nursery tank 3	Gill	Negative
				Mucus	Negative
				Skin	Negative
22	I39/1/22	4.0	Nursery tank 3	Gill	Negative
				Mucus	Negative
				Skin	Negative
23	I53/1/22	4.2	Nursery tank 3	Gill	Negative
				Mucus	Negative
				Skin	Negative
24	I54/1/22	5.4	Nursery tank 3	Gill	Negative
				Mucus	Negative
				Skin	Negative
25	I55/1/22	4.3	Nursery tank 3	Gill	Negative
				Mucus	Negative
				Skin	Negative

Figure 1. *Benedenia* sp., the parasite found on the skin surface of star pomfret (magnification 4000x).

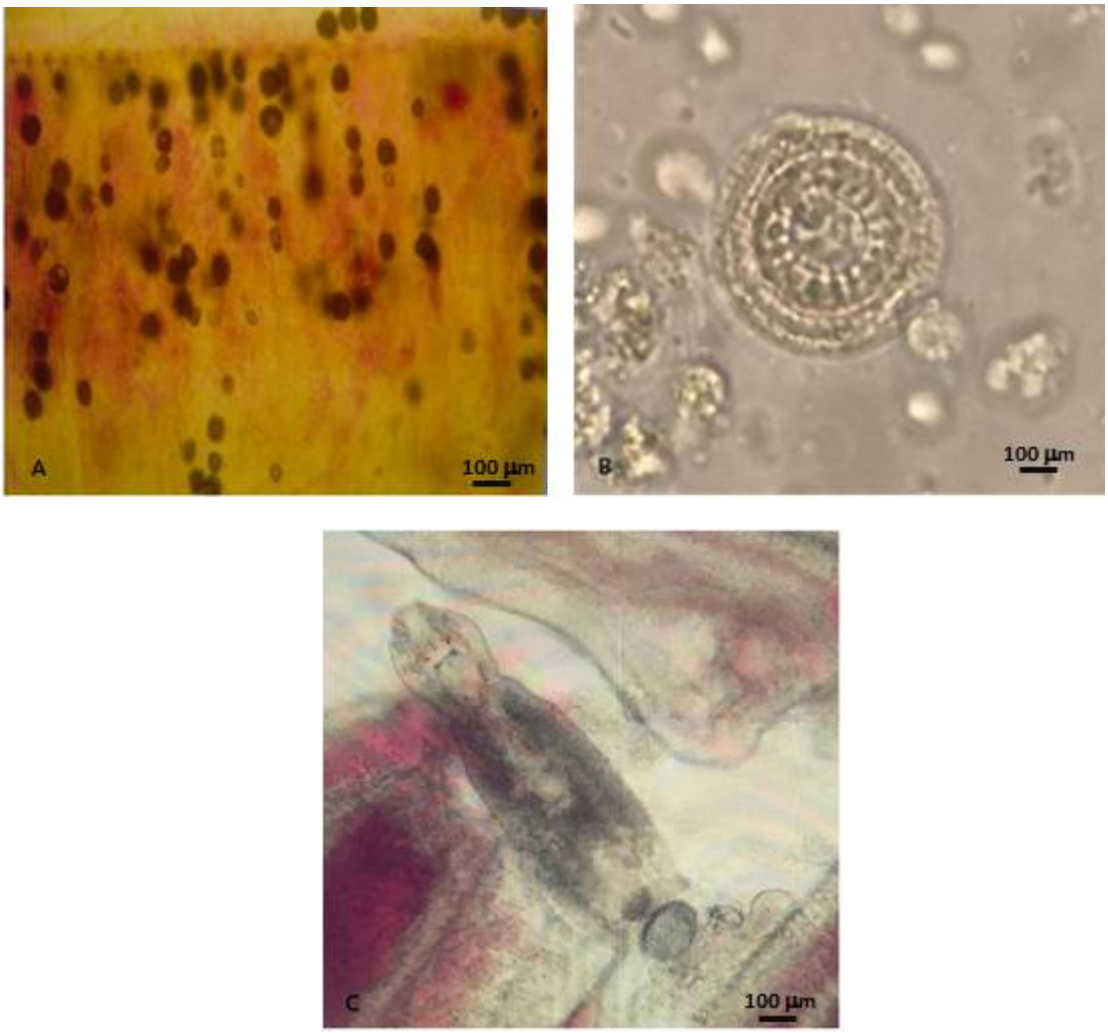

Figure 2. *Amyloodinium* sp. (A), *Trichodina* sp. (B) and *Diplectanum* sp. (C), the parasites found in the gills of star pomfret (magnification 4000x).

Discussion. This study found the parasites *Benedenia* sp. (Phylum: Platyhelminthes), *Amyloodinium* sp. (Phylum: Myzozoa), *Trichodina* sp. (Phylum: Ciliophora) and *Diplectanum* sp. (Phylum: Platyhelminthes) on the skin and gills of fish. The existence of these 4 genera is of course closely related to the conditions and cultivation system implemented at BMC, Batam. Parasite attacks generally only occur in large fish kept in FNCs that are in open sea waters. The stocking density of fish in FNC is much higher when compared to conditions in the wild. Such a stocking density provides ideal conditions for the spread of parasites, where each individual fish is relatively close together, and often even touches one another so that the parasites can easily move and spread from one individual to another. The water in FNCs is seawater from the open nature which of course is the habitat of ectoparasites. Examination for the presence of parasites is relatively rare, which takes place only once in 1-2 weeks. Fish that were heavily infested or even dead were not immediately removed and secured, instead, they are left in the FNC for a long time. This condition certainly worsens the conditions for handling and controlling ectoparasites in this place. In addition, fresh fish and trash fish as the main feed for this pompret can also be a source of ectoparasites.

Azuar et al (2019) reported that the parasites of star pomfret kept in FNCs around Bintan Island, neighboring Batam Island, were dominated by 3 genera, namely *Benedenia*, *Cirolana*, and *Zeylanicobdella*. It was further mentioned that the most dominant ectoparasite was *Zeylanicobdella arugamensis*. A somewhat different result was also reported by Hakim et al (2019) where the identified species of endoparasites that were successfully isolated in pomfret in Tanjung Pinang, Bintan Island, Indonesia were *D. varicus*, *Lecithochirium* sp., *Gorgonorhynchus* sp., *Ornithonyssus* sp. and *Camallanus* sp..

Benedenia sp. is included in the Monogenea class, the order Capsalidea and the family Capsalidae. This parasite has a flat body shape and has a pair of suckers on the anterior part of the body and a rounded opisthaptor on the posterior part of the body with an average diameter of 0.19 mm (Kabata 1985; Yang et al 2007). Jithendran et al (2005) added that the parasite has a body size of 2.05-3.29 mm length and 0.66-1.33 mm thickness and has two pairs of meta spots on the anterior and posterior parts. The anterior eye spots are smaller than the posterior. This parasite is an ectoparasite which is generally found on the skin, eyes, nasal cavity and gills. *Benedenia* sp. infection causes stunted fish growth, skin wounds, and secondary infection by bacteria. This parasite eats / sucks blood so that the fish looks thin and can cause blindness if the eye is the place of attachment. One of the results of this parasitic infection is that it causes sores that become an entry point for bacteria. Severe mortality due to this parasite can reach 30% (Ogawa et al 2020). The recommended treatment of diseases caused by *Benedenia* sp. is by dipping the fish in formalin solution (250 ppm) for 1 hour (Ogawa & Yokoyama 1998; Wiyatno et al 2012). This parasite infestation will result in reduced fish appetite, wounds on the skin surface and damage to the gill epithelium. Severe infestations will cause death in fish if present in large numbers (Sitjà-Bobadilla et al 2021).

Amyloodinium sp. is a parasite of the Oodiniaceae family in the dinoflagellates group. This parasite has a body size of 0.002-0.1 mm, while in the adult tropon phase it can reach a diameter of 120 m. It causes a disease called amyloodiniasis or velvet disease. This parasite is attached to the host tissue on the skin and gills using a short stalk or peduncle and at the end there are rhizoids and stomopods like tentacles that can move (Kumar et al 2015). Characteristics of fish infected with this parasite include restless, operculum slightly open, folded fins, and thinner body. Sometimes there are symptoms of fish often rubbing their bodies against hard objects such as tub walls, aerator stones and others. This parasite can cause 100% death within a few days (Noga & Levy 2006). At the Marine Cultivation Fisheries Center, Batam, it was reported that fish exposed to this parasite experienced mortality of up to 80% after being kept for 25 days. Its life cycle lasts 7-10 days. In early development, the larvae will stick to the skin and gills of fish. This larval form is difficult to see with the naked eye. After a few days of attachment, these larvae will become adults called trophonts and will produce up to 256 cysts (dinospora). These cysts will turn into tomites that can swim freely in search of new

hosts or fish. This tomite will turn into a trophont phase if it gets a new host. Tomites are able to survive for 1-2 days outside the fish's body before getting a new host. If it cannot find a new host it will die. Dinospores will continue to be produced and have an infectiousness that will occur at a temperature of 16 to 30^{o}C and a salinity of 10 to 60 ppt (Dequito et al 2015; Virgula et al 2017; Sitjà-Bobadilla et al 2021).

Trichodina sp. has a round shape when viewed from above and disc-shaped when viewed from the side. At the edge of the parasite it has cilia to move with hook-like denticles to attach to the host. This parasite infects the skin, fins and gills of fish. This parasitic infection causes the fish's body to be pale, thin, restless, sluggish, decreased appetite, damaged fins, excess mucus production and irritation of skin epithelial cells. The life cycle of *Trichodina* sp. occurs by binary fission. Cleavage occurs in the denticle of the parent cell which produces a new parasite. After this parasite attaches it will spin around and eat damaged epithelial cells (Yang et al 2007; Martins & Ghiraldelli 2008; Sufardin et al 2021).

Diplectanum sp. parasites belong to the order Dactylogyridea, family Diplectanidae and are known as monogenetic gill trematodes parasites. This parasite is also called gill worm; it is a parasite that is quite dangerous and is often found in marine fish. Some species of gill parasites can cause serious death in fish. It has a characteristic that distinguishes it from other species in the order Dactylogyridea, namely having a squamodisc (one on the ventral and one on the dorsal), and a pair of anchors that are located far apart (Kabata 1985; Yang et al 2007; Musman et al 2015). This parasite attacks the gills of fish causing the gills look pale or swollen. The body of fish infected with this parasite looks pale, thin, sluggish, and has reduced appetite. Other symptoms can also be increased respiratory frequency and excess mucus production on the gills. Fish that are infected by the parasites often gather on the surface or in the water inlet in search of oxygen. *Diplectanum* sp. has a direct life cycle, meaning that it does not involve an intermediate host. The life cycle starts from eggs released in the waters, then 2-3 days will form ciliated larvae (oncomiracidium). Oncomiracidium move freely in nature (waters) for 6-8 hours up to 24 hours, then look for the right host. Oncomiracidium will attach to the gills and develop into adults (Grabda-Kazubska & Kiseliene 1991; Martins & Ghiraldelli 2008; Sitjà-Bobadilla et al 2021).

Conclusions. In this study the following parasites were identified, namely *Benedenia* sp. (Phylum: Platyhelminthes), *Amyloodinium* sp. (Phylum: Myzozoa), *Trichodina* sp. (Phylum: Ciliophora), *Diplectanum* sp. (Phylum: Platyhelminthes) on the skin and gills of star pomfret. While in the mucus, no parasites have been found so far. Parasitic disturbances are closely related to water quality management and aquaculture management globally. Therefore it is recommended for improvement on these sides.

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Conflict of interest. The authors declare that there is no conflict of interest.

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