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Parasitic Protozoa found in the skin, gills, and intestines of Patin Catfish (*Pangasius hypophthalmus*) and Common Carp (*Cyprinus carpio*)

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Abstract

The parasitic protozoa found in "Ikan Patin" or a species of Catfish (*Pangasius hypophthalmus*) and "Ikan Mas" or Common Carp (*Cyprinus carpio*) are *Trichodina sp.*, *Ichthyophthirius sp.* and *Myxobolus sp*. These protozoa have the potential to cause death in fish and economic losses to fish farmers and sellers. This study was conducted to determine the presence of parasitic protozoa on the skin, gills, and intestines of Catfish and Carp. Samples were taken from the Bursa Ikan Hias Laladon or Laladon Ornamental Fish Market in Bogor, Indonesia. Each species was sampled for as many as 30 fish. The skin, gills, and intestines were examined using the native examination method and Lugol staining. Parasitic protozoa were identified based on their morphology, namely size, and shape. The types of parasitic protozoa found on the skin, gills, and intestines of Catfish and goldfish were *Trichodina* sp., *Ichthyophthirius* sp., and *Myxobolus* sp. The parasites were found in skin organs in as many as 29 samples (96.7%) from Catfish and 30 samples (100%) from Carp. The Genera of *Ichthyophthirius* sp. were found in the gills of 2 samples (6.7%) in Catfish and 1 sample (3.3%) from positive Carp. *Myxobolus* sp. was found in the intestines of 11 samples (36.7%) from Catfish and two samples (6.7%) from Carp. The species of *Trichodina* sp. found based on morphology was *Trichodina giurusi*.

Keywords: *Cyprinus carpio*; *Pangasius hypophthalmus*; patin; parasitic Protozoa; *Trichodina* sp.

1. Introduction

The fisheries sector plays an essential role in providing animal protein for the people of Indonesia. Fish production reaches approximately 2 million tons per year, most of fish production (74%) comes from the sea, and the rest (26%) comes from freshwater [1]. Fish is a source of animal protein that is high in protein and easily digested by the body and can meet the nutritional needs of the Indonesian people [2]. Therefore, the need for animal protein sources from fish is increasing. Increased production is also accompanied by the spread of disease in fish. Today, fish farmers often complain of disease problems in fish, especially freshwater fish [3]. Internal and external factors that play a role in the onset of disease. Internal factors come from the environment in the fish's body, such as genetic disorders [19], immunity [20], and body metabolism. Pathogenic agents influence external factors, namely parasites, viruses, fungi, and bacteria. The emergence of infection with non-pathogenic agents is caused by temperature, water quality, pH, toxic gases, and nutrients [4]. Disease in fish is one of the problems often encountered in fish farming. The presence of fish disease is closely related to the management of the fish's habitat [5].

The commodity value of Catfish (*Pangasius hypophthalmus*) and Carp (*Cyprinus carpio*), apart from being used as fish for consumption, they can also be used as ornamental fish, especially small ones. Freshwater fish farming, especially Catfish and Carp, is currently facing fish disease problems. This problem causes a decrease in production, a decrease in fish quality, and even total death, thus causing economic losses to freshwater fish farming farmers [6]. Globally, the potential for economic losses due to disease outbreaks caused by infection with pathogenic microorganisms shows a significant loss. Economic losses to the aquaculture industry due to disease outbreaks are estimated at US$ 9 billion per year and have an impact on decreasing the number of aquaculture production worldwide [7].

Diseases that are often found in freshwater fish are trichodiniasis, myxosporidiasis, and ichthyophthiriasis [8]. All three diseases are caused by protozoa that cause damage to the gills, intestines, and skin of fish. Trichodiniasis is caused by the parasite *Trichodina* sp. and is often found on the gills. *Myxobolus* sp. causes myxosporidiasis [9]; this parasite is commonly found in the digestive organs of fish. Meanwhile, ichthyophthiriasis is a disease caused by the parasite *Ichthyophthirius* sp. [10, 11]. These parasites are found in freshwater fish's epithelial skin and mucous membranes (ornamental fish and fish consumption). This disease is known as ich or white spot in fish. Therefore, it is necessary to conduct research to identify parasitic protozoa found on the skin, gills, and digestive tract of Catfish and Carp. This study aims to identify parasitic protozoa found on the skin, gills, and digestive tract of Catfish and Carp.

2. Experiments Procedure

Sample Collection

Fish samples were obtained from the Laladon Ornamental Fish Market, Bogor. Samples were taken randomly from as many as 60 fish consisting of 30 Catfish with a bodyweight of ± 5 grams and 30 goldfish with a bodyweight of ± 5 grams, then brought to the Protozoology Laboratory for examination of parasites in fish.

Protozoa Examination and Identification Method

Parasite examination was carried out on several parts of the fish's body, namely skin scrapings, gills, and digestive tract. The sampling procedure is as follows:

a. Skin scraping

Examination of parasites attached to the body's surface is carried out by scraping using a razor from the cranial to the caudal direction carefully in the superficial part. Next, skin scrapings are smeared on the object's glass.

b. Gills examination

The gills are examined by taking the gill sheet directly with scissors; then, it is reviewed on an object-glass.

c. Digestive tract examination

The intestinal examination is done by slicing the intestines and removing the contents of the intestines, then reviewing them on an object-glass. The examination and identification methods were carried out by two methods, namely native examination and Lugol staining. The native examination was carried out by adding one drop of distilled water in the object-glass on the organ scrapings (skin, gills, and intestines) of the fish being examined and covered with a cover glass. For Lugol staining, the object-glass was given one drop of distilled water and one drop of 1% Lugol. After that, skin, gill, and intestinal scrapings were taken and reviewed as in the previous procedure. The preparations were observed with a microscope in a zigzag manner at magnifications of 100 times and 400 times.

Counting the number of Protozoa

The number of protozoa was calculated by counting all the protozoa observed in 20 fields of view. Calculation of the number and identification of the type of protozoa carried out in each field of view.

3. Result and Discussion

The identification results in Catfish and Carp. The parasitic protozoa found on the scales or skin, gills, and intestines were: *Trichodina* sp., *Ichthyophthirius* sp. dan *Myxobolus* sp.

Trichodina sp.

The identification results showed that as many as 29 samples (96.7%) of Catfish were positively infected with *Trichodina* sp. on skin organs, and 19 samples (63.3%) were positively infected on the gills. In addition, *Trichodina* sp. is also found in the skin, gills, and intestines of the Carp. The highest infection was found in skin organs; as many as 30 samples (100%) and 29 samples (96.7%) were positively infected on the gills (Table 1).

Table 1. The results of the examination of *Trichodina* sp. on Catfish and Carp

Fish Name	Quantity	Positively Infected Examination Results (%)		
		Skin	Gills	Intestine
Patin	30	29(96.7)	19(63.3)	12(40)
Mas	30	30(100)	29(96.7)	3(10)

According to [12], *Trichodina* sp. can infect fish, especially on the gills and skin. This is because *Trichodina* sp. has denticles that can stick to fish organs so that they can infect fish. *Trichodina* sp. has denticles or hooks teeth on the body's surface; it is round like a disc. *Trichodina* sp. comes from the Ciliata class, which has cilia or vibrating hair on the surface of its body. Generally, healthy fish are able to control the number of parasites in their bodies. However, in very large numbers, *Trichodina* sp. is able to lower the immune system and open the path of infection for other organisms, such as bacteria.

The high factor of *Trichodina* sp. is allegedly due to poor maintenance management at the Laladon Ornamental Fish Market. One of them is the frequency of water changes that are not regular, and the density of fish in the aquarium is not paid attention to. *Trichodina* sp. will eat bacteria on the surface of the fish body to stimulate the reproduction of *Trichodina* sp. by cell division. According to [13], trichodiniasis is caused by less-than-optimal environmental conditions, high fish density, pond management, and poor feed.

The incidence of trichodiniasis in Carp is higher than in Catfish. One of them is because the body structure of the Carp has scales so that the denticles of *Trichodina* sp. are easily attached to fish organs compared to Catfish, which have slimy skin. According to research conducted [14], *Trichodina* sp. is also found in the skin and gills of Tawes fish (*Puntius javanicus*).

Types of *Trichodina* sp. what was found was *Trichodina giurusi*. This species has a diameter range between 24.4-34.8 m, as shown in Tables 2 and 3. *Trichodina giurusi* is classified as *Trichodina* sp. small ones [15]. *Trichodina* sp.

small sizes have higher pathogenicity than larger ones [16].

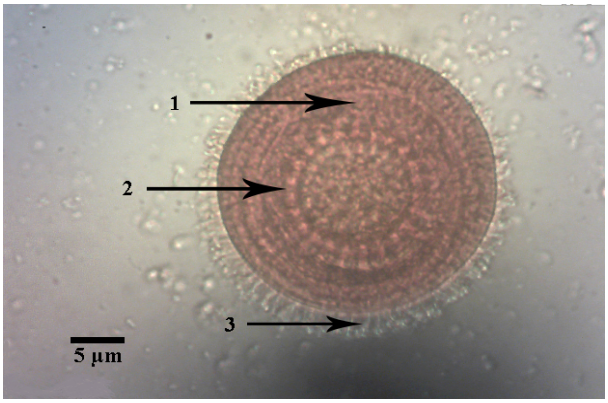

Figure 1. Morphology of *Trichodina* sp. with a magnification of 400 times. Description: 1. Radial pin 2. Denticle 3. Cilia

Table 2. Characteristics of *Trichodina giurusi* in Catfish, Carp, and Butini fish

Characteristics	Size Range	Size Range	Size Range
Diameter (µm)	16.96 - 36.07 (24.6 ± 3.5)	14.37 - 41.01 (23.6 ± 4.0)	24.4 - 34.8 (29.7 ± 2.7)
T.giurusi Number	297	769	20
Host	Patin Catfish	Carp	Butini Fish
Country	Indonesia	Indonesia	India
Reference	Research	Research	Mitra/Partner (2005)

Table 3. Characteristics of *Trichodina giurusi* in Catfish and Carp by organ.

Characteristics	Organ	Patin Catfish	Carp
Diameter (µm) Range [Quantity of *T.giurusi*]	Skin	17.31 – 34.51 (24.5 ± 3.4) [236]	14.37 – 41.01 (24.5 ± 4.0) [560]
Diameter (µm) Range [Quantity of *T.giurusi*]	Gills	17.11 – 30.35 (24.5 ± 3.3) [43]	14.61 – 29.13 (21.0 ± 2.5) [203]
Diameter (µm) Range [Quantity of *T.giurusi*]	Intestine	16.96 – 36.07 (25.3 ± 4.9) [18]	17.19 – 24.08 (20.7 ± 2.8) [6]

Ichthyophthirius sp.

The results showed that infection with *Ichthyophthirius* sp. was found only in the gills of Catfish; as many as two samples (6.7%) and 1 sample of goldfish (3.3%) were positively infected, as can be seen in Table 4. *Ichthyophthirius* sp. is a protozoan that can attack the skin and gills of fish in a relatively short time and comes from the Ciliata class. Unfavorable environmental conditions such as poor sanitation of water ponds, unsuitable water temperature, and salinity, in addition to weak immune systems in fish, so that fish become more easily stressed and infected with *Ichthyophthirius* sp.

Table 4. Examination results on *Ichthyophthirius* sp. in Catfish and Carp.

Fish Name	Quantity	Positively Infected Examination Results (%)		
		Skin	Gills	Intestine
Patin Fish	30	0(0)	2(6.7)	0(0)
Carp	30	0(0)	1(3.3)	0(0)

According to [10], *Ichthyophthirius* sp. attacks by sticking to the mucus layer and sucking red blood cells. However, skin and gut samples from both fish were negative for *Ichthyophthirius* sp., which can be caused by fish diet, fish resistance, and fish environmental conditions. According to research conducted by [17], *Ichthyophthirius* sp. is found in skin scrapings and gills of Catfish.

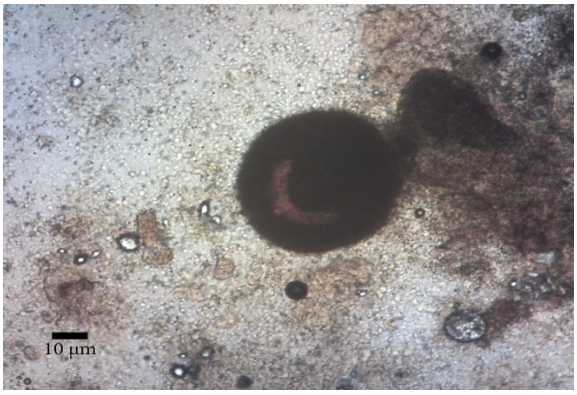

Figure 2. Morphology of *Ichthyophthirius* sp. with 400 times magnification

Myxobolus sp.

Based on Table 5, 11 samples (36.7%) of Catfish were positively infected with *Myxobolus* sp. in the intestine, and 2 samples (6.7%) of goldfish were positively infected with *Myxobolus* sp. in the intestines.

Table 5. The results of the examination of *Myxobolus* sp. on Catfish and Carp.

Fish Name	Quantity	Positively Infected Examination Results (%)		
		Skin	Gills	Intestine
Patin Catfish	30	0(0)	0(0)	11(36.7)
Carp	30	0(0)	0(0)	2(6.7)

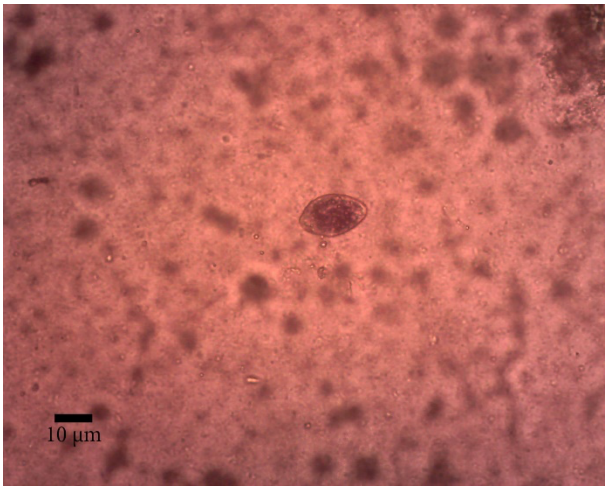

Figure 3. Morphology of *Myxobolus* sp. with 400 times magnification

Myxobolus sp. from the class sporozoa. Catfish are more susceptible to *Myxobolus* sp. because of a different diet than goldfish. Goldfish are omnivores or mixed eaters, so the gut flora is better than Catfish, which tend to be carnivores or meat-eaters.

Myxobolus sp. can cause considerable economic losses because it causes a decrease in nutrient absorption from feed and growth disorders. According to [18], *Myxobolus* sp. can be transmitted through the feces of infected fish and then ingested by healthy fish. Skin and gill samples on both fish showed negative for *Myxobolus* sp. This is in accordance with [13] that the habitat of *Myxobolus* sp. is found in the fish's digestive tract (gut). According to research conducted by [6], *Myxobolus* sp. is also found in the intestines of Betutu fish (*Oxyeleotris marmorata*).

4. Conclusion

The examination results showed that the types of protozoa found in Catfish (*Pangasius hypophthalmus*) and Carp (*Cyprinus carpio*) were *Trichodina* sp. and *Ichthyophthirius* sp., which includes ciliates and *Myxobolus* sp. which includes sporozoans.

The most common protozoa found on the skin and gills of Catfish (*Pangasius hypophthalmus*) and goldfish (*Cyprinus carpio*) were *Trichodina* sp. (ciliate), while the most common protozoa found in the intestines is *Myxobolus* sp. (Sporozoa).

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