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Case Report

New Report on Trichodiniasis (Protozoa: Ciliophora: Peritrichida) in Jade Perch; *Scortum barcoo* from Peninsular Malaysia

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Abstract

Background: Infestations caused by trichodinids are particularly significant in aquaculture because they give serious impact and responsible for causing problem in growth, decreased immune response in vaccinated fish, chronic mortality during production and changes in vision and swimming in larvae and culminating in acute mortality. **Objective:** This study purposely to report the trichodinid parasites infested the new commercial cultured species; *Scortum barcoo* (Jade Perch) for aquaculture industry in Malaysia. The occurrence of Trichodinid parasites infestation were detected after five days upon arriving of the fish. **Materials and Methods:** Ten samples of *Scortum barcoo* were collected from tank-cultured at Sekayu, Kuala Berang, Terengganu, Malaysia (4°57'57.9"N, 102°57'25.9"E). Sample's weight and length were measured, respectively, then were diagnosis directly for ectoparasites. The average weight and length of samples collected are 736.4 ± 89.6 g and 46.6 ± 1.6 cm, respectively. **Results:** Clinical symptoms of the infestation showed are red lesion on the body, the darkening of body colour, fish exhibited flashing behaviour on the wall and bottom of the tank. Parasites collected was study by observing and drawing of their morphological characteristics. The important parts of the parasites organ were identified for further identifications i.e., denticle which consist denticular ring, blade, cilia, etc. The record of water quality were taken to relate the symptom of the infestations. **Conclusion:** This study assumed that the inhibiting of this ectoparasites are related to the handling procedures of fish during transportations. Improper acclimatizing procedure during transportations may contribute to the spread of this ectoparasites. Saltwater bath were applied to prevent this parasite from spread. This observation's study was first report for trichodinid parasites infestation on the Jade Perch; *Scortum barcoo* at the east coast of Peninsular Malaysia.

Key words: Trichodinid, Jade Perch, *Scortum barcoo*, *barcoo* grunter, Peninsular Malaysia

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

*Scortum barcoo*¹ is a species of fish under family of Terapontidae and known as Barcoo Grunter or Jade Perch. It is endemic to Australia, where it can be found in certain major rivers, including the river basins of Australia with the Lake Eyre Basin² and Barcoo river. It is commonly reared in hatcheries for commercial aquaculture industry. This fish morphology has a sturdy body and a small head. The colour of body is greenish to brownish with darken blotches at one side of fish body and darker fins². The fish reaches about 35 cm in maximum length. This species is omnivorous which eating crustaceans, insects, molluscs and fish are they mains prey³. Nevertheless, this species is newly introduced to Malaysia for future commercial aquaculture production. There is no report or study have been published regarding for this species of fish in Malaysia. This species also have a potential to spread and also infested by surrounding diseases.

Trichodiniasis is ubiquitous parasitic disease caused by protozoans of the family Urceolariidae that infects various cultured and wild fish. Members of this family are commonly referred as trichodinids and are divided into various genera, the most common of genus which are *Trichodina*, *Paratrachodina*, *Tripartiella*, *Trichodinella* and *Dipartiella*. Worldwide, over 270 species of trichodinids have been described according to Valladao *et al.*⁴, but most of studies were conducted on their relationship with the host in captivity. Infestations caused by trichodinids are particularly significant in aquaculture because they are responsible for causing decreased growth, a decreased immune response in vaccinated fish, chronic mortality during production and changes in vision and swimming in larvae and culminating in acute mortality^{4,5}. There is more addressing the symptom pathology associated with this parasitic infestation⁶⁻⁸. The main changes that have been described related to symptom of muscle and tissue damage such as hyperplasia, hypertrophy, oedema, inflammatory infiltration, muscle necrosis etc.^{9,10}. The main protozoan parasites that commonly infested the fish are *Trichodina* sp. This species morphology are dorsal-ventrally flattened oval ciliated protozoan parasites. Common species that can found on marine and freshwater species of finfish^{5,11,12}. A readily distinguishable characteristic of these organisms is the presence of a prominent denticle or "Blade-like" internal cytoskeleton ring called denticular ring. There are four additional genera of trichodinid which are similar in description and life cycle¹³. While, small numbers of these organisms on a fish generally do not cause much of a

health problem, large numbers can cause moderate to serious pathology and ultimately caused high mortality of fish. Small fish and juvenile are especially susceptible and mortality can occur quickly if undiagnosed earlier⁶.

Now a days, much study had been done regarding parasitic disease treatments. Fish farmers have used disinfectants such as formaldehyde for the control of parasitic diseases¹⁴. Despite, the use of these substances is permitted in some regions, such as North America¹⁵. However, in other country (illegal), these substances are even used without proper monitoring and can caused the environment pollution. Hence, the effectiveness of some treatments is not fully describes. To date, there is no report of trichodiniasis infestations on Jade Perch in Malaysia. Previous studies more stressed on study of ectoparasites from Silver perch mostly from Australia^{16,17}. There is a report of *Trichodina* sp., in a wild fish such as study that have been done by Ihwan *et al.*¹². This group of parasites are categorized as ectoparasites which consume the nutrient from the host and caused the decrease of the immune resistance of the host.

MATERIALS AND METHODS

Sample collection: Ten samples of *Scrotum barcoo* were collected from hatchery at Sekayu, Kuala Berang, Terengganu, Malaysia (4°57'57.2"N, 102°57'26.8"E). Measurement of length and weight were recorded and directly diagnosed for ectoparasites by following standard methods¹². Data for the samples collected with average weight 736.4 ± 89.6 g and length 46.6 ± 1.6 cm can be referred to Table 1. The samples of parasites were collected from the body and gill using smear techniques. All experiments were done in triplicates¹⁸. The number of ectoparasites were count for each replicate as maximally 15 cells; or simply stated as more than 15 cells per smear (>15). The selected of ectoparasites were observed and drawn for the morphological identifications. Standard procedure for freshwater parasites study were referred to Fernando *et al.*¹⁹. Study was done in Laboratory of Aquatic Organism Health (AOHU), Institute of Tropical Aquaculture (AKUATROP) and corporation with private commercial hatchery.

Table 1: Average weight and length for samples of 10 fish (*Scrotum barcoo*) collected

Samples	Weight (g)	Length (cm)
Mean	736.4	46.6
Standard deviation	89.6	1.6

RESULTS AND DISCUSSION

The morphological identifications of ectoparasites showed *Trichodina* sp., were recorded infest on *Scrotum barcoo*. Despite, *Trichodina* sp., is one of the ectoparasites that were found in this study (Fig. 1). This parasites species was infested the fish host in higher of intensity (>15-30 individuals per replicates of slide smear) with the prevalence of 100% (Table 2) on the body and gills of the host. Study done by Valladao *et al.*⁴ showed the suction areas on the larval integument and desquamation. Normally, ciliates have a medium size bell-shaped body, adhesive disc, denticles ring with the number of 21 denticles, adoral cilia and the organ of reproductive (Fig. 2). Specimens analysed in this study exhibit some morphological variation with respect to the shape of the body.

Inspite of, this species of parasites are well-diverse which is recorded from varied host fish species, mainly cyprinids around the world²⁰. According to Ihwan *et al.*¹², this species also present in the wild environment. Furthermore, this species are ubiquitous and probably attained via transcontinental introductions of the host, mainly fish.

Particularly, this species outbreak starting from the improper quarantine procedures. This species has been dispersed to other locations together with the poor handling and transportations¹. Nevertheless, it possess a large dispersal capability and now a days it is now not only found in introduced hosts but also in the native freshwater fish fauna. *Trichodina* sp., can cause extensive fish mortality in an aquaculture system¹³. The ability of this parasite to quickly multiply under certain environmental conditions or when the fish are stressed by other factors makes early detection of this parasite a high priority in an aquaculture facility. Early

Table 2: Data for fish samples infested by trichodinid parasites

Samples	Replicate 1	Replicate 2	Replicate 3
Fish 1	10	8	>15
Fish 2	3	9	5
Fish 3	>15	13	>15
Fish 4	13	14	10
Fish 5	3	>15	>15
Fish 6	>15	>15	>15
Fish 7	>15	11	6
Fish 8	13	>15	>15
Fish 9	>15	>15	>15
Fish 10	>15	14	13

*Prevalence of infestation are 100%

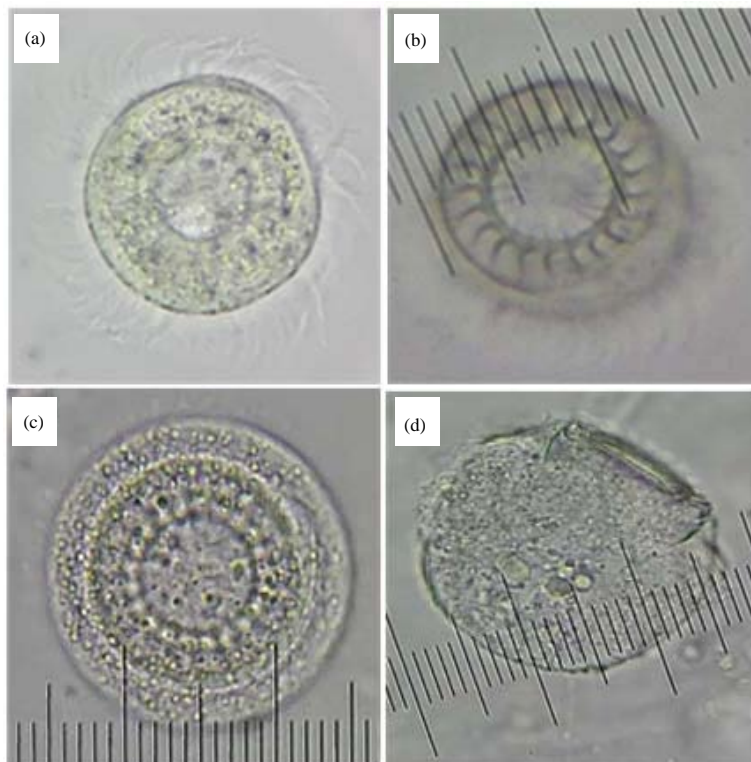


Fig. 1(a-d): Micrograph of *Trichodina* sp., (a) Structure of cilia, (b) Denticular ring, (c) Posterior view and (d) Side view of the ectoparasite (400x magnification)

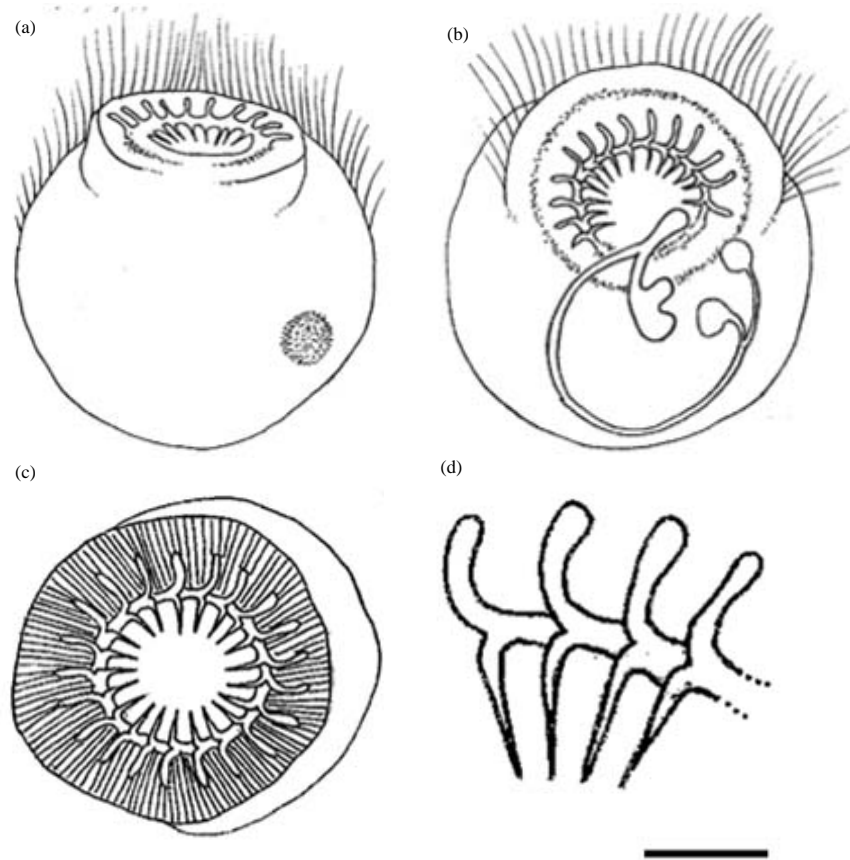


Fig.2(a-d): Illustration drawing of *Trichodina* sp., (a) Whole cells of parasites, (b) Structure of reproduction system, (c) Denticular ring and (d) Denticle with the shape of blade (Compound Microscope-Nikon Eclipse 200 attached with Nikon Lucida camera Y-IDT, Japan), scale bar: 1 μ m

detections can prevent the high mortality of the fish. An appropriate treatment or management response is essential to stop the outbreaks. A common treatment procedures and other related recommendations are discussed in this report.

Symptom of infestation: *Trichodina* sp., can cause irritation by feeding on the epithelial layer of cells covering the surface of the gills and skin of the fish. This can result in hyperplasia (proliferation) of the epithelial cells, clubbing of the gill filaments and even fusion of the gill filaments¹³. This affects the ability of the gills to maintain optimal respiratory and excretory activities and the ability of the skin to maintain proper homeostatic osmoregulatory properties. Massive infestations of these parasites on fish can also directly result in superficial to deep ulcerative skin lesions which then allow for secondary bacterial and fungal infections to develop at the affected site⁴. From this study, fish infested by this ecto-parasites show some clinical symptom i.e., red lesion on

body, fin root, exophthalmos, darken body colour, dislodged scales, etc. Other symptom that showed by the infested fish are flashing on the wall and bottom of the tank, whirling and also dyspnoea. Figure 3 shows the physical symptoms of infestation during the parasite occurrences.

Other parasite infestations: From the observation, there is other parasites were found such as monogenea (*Gyrodactylus* sp.) and fungus but *Trichodina* sp., show the highest numbers of infestation which >15 number of parasites per replicate smear. The picture of other parasites infestation can be referred to Fig. 4.

Water quality: In general, this type of parasites are related to the drastic temperatures changeability. However, in this study, there is no significant relationship between the parasites and the physical parameter. Water temperature data are considerably normal and this can be referred to the Table 3.

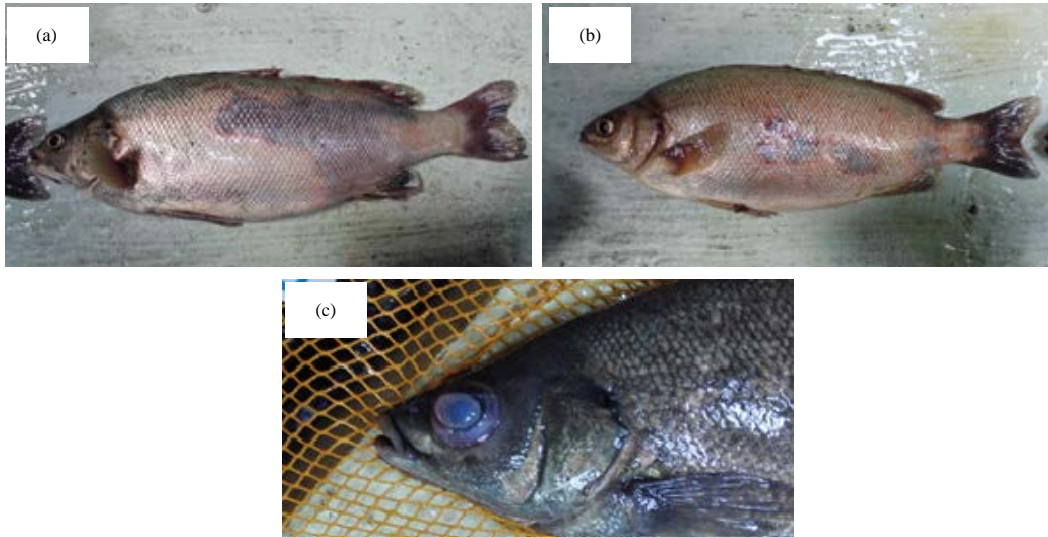


Fig.3(a-c): Symptoms of infestation during the study, (a) Fin root+lesion, (b) Red lesions and (c) Exophthalmos, (Pop-eye)+changes of body colour

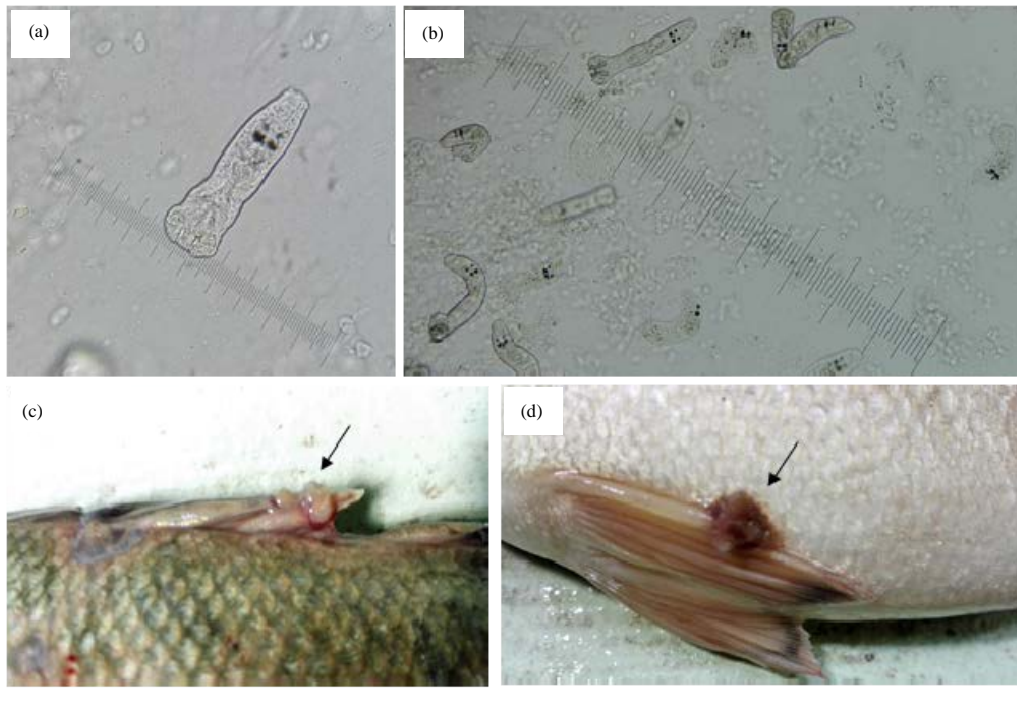


Fig. 4(a-d): Other parasites that found during the study photo, (a) *Gyrodactylus* sp., (b) Monogenea on the body of the fish (400x magnification), (c) Dorsal fin and (d) Anal fin the infestation of fungus (arrow) on the fin (Nikon Eclipse E200)

Table 3: Physical and chemical parameter that recorded during the study

DO (mg L ⁻¹)	pH	Temperature (°C)	Turbidity	NH ₃	NO ₂ ⁻	PO ₄
5.80	6.59	27.47	8.62	0.309	1.841	0.003

The problem is chemical parameter especially results for ammonia (NH₃), nitrite (NO₂⁻) and phosphate (PO₄) shows a

slightly high concentration and shall consider as one of the contribution factor of infestations.

Prophylaxis and treatment: Currently, the effectiveness of a treatment protocol and the potential to minimize the use of

chemicals were evaluated. This can decrease the long term effects of the fish and human consumption. There are several methods by which *Trichodina* sp., may be controlled in the aquaculture. These include the usage of chemical treatments, saltwater baths and flushing. The UV light treatment is generally considered ineffective due to the high dosage rates required to kill the organisms²¹. This study applied the saltwater bath as a conventional methods which is more safe and cheaper. From the previous studies, the fish were treated using saltwater (NaCl) solution bath with concentration ranged between 3-6 ppt for a week treatment¹⁶. This study were modified the salt concentration volume which dilution applied 10-15 ppt of saltwater bath for 3-4 h dip.

CONCLUSION AND FUTURE RECOMMENDATIONS

Symptoms of Trichodinid infestation on Jade Perch, *Scortum barcoo* were clearly showed during the observations. This study can be a pioneer reports of *Trichodina* sp., occurrences at the east coast of Peninsular Malaysia. The results for water quality shows that there is slightly significant relationship to relate the symptom of the infestations. This study expected that the inhibiting of this ectoparasites are related to the handling procedures of fish during transportations. Improper acclimatizing procedure during transportations may contribute to the spread of this ectoparasites. Saltwater bath were applied to prevent this parasite from spread. Further DNA study can be done to fully identify the species of this *Trichodina* sp.

Mortalities in moderated infested fish by *Trichodina* sp., is usually associated with one or more of stress factors including rough handling, overcrowdings, malnutrition, high ammonia as well as low oxygen. Temperature, pH and salinity involved when acclimatizing aquatic life to the new aquatic environment. Once the fish in the bags/tank are transferred, the high-pressure gradient of oxygen in water will be supplied. Fish that acclimatize too quickly can include the sign and symptoms i.e., rapid gill movement; pectoral and dorsal fins held rigid, pale coloration, loss of orientation, etc. Correct quarantine procedures may further compromise fish survival. Furthermore, quarantine procedures is to ensure that the transportation is clear of transmittable ectoparasites; before and after its release into the new environment. It was involves the holding of fish under controlled condition in tanks over an approved period and checks for diseases. Life fish should be not fed 12-24 h before transporting. The reason of fasting the fish is to prevent the high-release of ammonia which may pollute the water and cause the fish mortality. As a precaution, it is advisable that isolation and observation of the new

arriving fish has to be practiced and hence this will maintain the immune system of the fish.

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