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Dian Eka Ramadhani[1*)], Farah Hasna Arafah[1)], Tetty Barunawati Siagian[2)], Mohamad Iqbal Kurniawinata[1)], Mad Rudi[3)], Rizky Fadilla Agustin Rangkuti[4)]

[1] Study Program of Technology and Management of Applied Aquaculture, College of Vocational Studies, IPB University, Bogor, Indonesia

[2] Study Program of Paramedic Veterinary, College of Vocational Studies, IPB University, Bogor, Indonesia

[3]Marine and Fisheries Education Study Program, Universitas Pendidikan Indonesia, Bandung, Indonesia, 40154

[4]Study Program of Aquaculture, Faculty of Fisheries and Marine Science, Brawijaya University, Malang, Indonesia, 40154

The Effect of Ectoparasites on Hatchery Business of Red Tilapia *Oreochromis* sp. in Klaten, Central Java

Corresponding author: dianeka06@apps.ipb.ac.id

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ABSTRACT

Tilapia fish hatchery in Klaten uses the Dengkeng River as a water source for fish hatchery. The polluted water quality of the Dengklek River increases the number of ectoparasites that can infest tilapia seeds. The purpose of this study was to evaluate the effect of ectoparasites on red tilapia in the Klaten area, Central Java. This research was conducted at the Freshwater Fish Seed Work Unit, Loka Janti, Klaten. The research sample used red tilapia seeds *Oreochromis niloticus* measuring ±4-6 cm. Fish sampling was carried out randomly at the Freshwater Fish Seeding Work Unit Loka Janti, Klaten. Sampling was performed on as many as 5 samples from 28 semi-permanent ponds. Physical and ectoparasite examinations were peerformed on tilapia seeds. The data analyzed included water quality, epidemiology, parasite count measurements, specific growth rates, financial values, and Sensitivity Analysis. Ectoparasites can affect tilapia cultivation and cause loss. Fortunately, the ectoparasites in Loka Janti did not cause any harm. Tilapia hatchery harvest size 2–3 cm with a selling price of Rp75.00 fish^{-1} resulted in an income of Rp426.062.70,00 and a profit of Rp71.814.554,00. The R/C ratio obtained was 1,20, and the payback period was 4,4 years.

Key words: red tilapia, ectoparasites, financial value, hatchery business,

1. INTRODUCTION

Nile tilapia *Oreochromis niloticus* were introduced to developing countries and cultured on a subsistence level to meet local protein needs (FAO 2022). In hatchery of Tilapia, the stage of larvae and fry is the critical point in fish farming. Good larvae and fry will produce good output as well. The availability of adequate fry quantity, quality and sustainability must be guaranteed, so that the fish farming business can run well.

Unfortunately, when the farmers increase the production with the super intensive method which is high density of fish, it leads to increase the disease attack in fish (Wang *et al.* 2023, Machimbirike *et al.* 2019; Surachetpong *et al.* 2020).

The disease attack can be caused by infectious (bacteria, virus, fungi, parasite and protozoa) and non-infectious disease (environmental condition, genetic, and nutrition). The fish skin and gills represent major pathways for pathogen agents to invade fish and thus act as a critical line of defence for maintaining host health by mucus layer (Glover *et al*. 2013; Lazado & Caipang 2014; Merrifeld & Rodiles 2015). Ectoparasites are disease agents that often infest fish in hatcheries. Ectoparasites are disease agents that often infest fish in hatcheries. The predilection of ectoparasites on the fish body is the skin and gills of fish. The skin and gills of fish are the main routes for pathogenic agents to infect fish. The skin and gills of fish act as a line of defence for the body to maintain the health of the host body against pathogenic agents such as ectoparasites through a layer of mucus (Depnath *et al.* 2022). Monogeneans are highly diverse fish ectoparasites with a direct life cycle, widely distributed and are known to generally display strict host specificity (Scheilfler *et al.* 2022). The most common symptoms of ectoparasitic infestations in tilapia fish fry are retarded growth rate, abnormal behaviour, epithelial lesions, blindness, deformities of gills, mass mortality, and consumer rejection (Tessema 2020; Misganaw 2016; Claude *et al*. 1998).

Ectoparasites that are often found in larvae and fry tilapia hatchery activities include *Dactylogyrus* sp., *Gyrodactylus* sp., and *Trichodina* sp. (FAO 2023; El-Sayed 2020). Those ectoparasites appears and can infect fish on the skin, gills and fins due to several factors including low water temperature, intensive culture, poor water quality and poor handling management (El-Sayed 2020). The presence of parasites eventually causes an economic loss for the fishing industry (Palm *et al.* 2008) and it is likely to be carrier state and do not always cause disease in fish (Barber 2007). Comparing to several business, this ectoparasites are also infected the juvenile common carp and pangasius fish as well as it leads to economic loss (Nematollahi *et al.* 2012; Ozan *et al.* 2008; Haque *et al.* 2004).

Tilapia hatchery business in the Klaten area is of concern because they use water sources from rivers. Utilization of river flow for tilapia hatchery activities will certainly be one of the determinants of the success of fish hatchery. The river flow that is utilized is the Dengkeng River which flows from upstream and downstream into the Bengawan Solo River with a length of approximately 45 km with an area of watershed of approximately 700,000 km^2. This river passes through one of the batik craft business centres in Kebon Village. One of the impacts of this business is that the resulting waste affects water quality. Several studies have reported that the water quality in the Dengkleng River has undergone water quality degradation from upstream to downstream. Based on the pollution load, this river has increased significantly with a biological oxygen demand (BOD) value of 25.57 kg day^{-1} and a chemical oxygen demand (COD) of 223.43 kg day^{-1} (Budianta *et al*. 2021). It is feared that the high value of these pollution will cause an increasing number of ectoparasites and other disease agents. Therefore, this study aims to evaluate the effect of ectoparasites on red tilapia in Klaten area, Central Java.

2. MATERIALS AND METHODS

2.1 Time and Location

This study was conducted from January to April 2022 at Working Unit of Fresh Water Fish Hatchery Loka Janti, Klaten, Polanharjo District, Central Java (Fig 1). The water source used for the fish hatchery comes from the Umbul Nilo and Umbul Wanut Rivers, which are 2 km from Loka Janti, where these water sources also in line into the Dengkeng Bengawan Solo River. In addition, the water sources is adjacent to irrigation canal of residents which are usually used for agricultural activities. The Umbul Nilo and Umbul Wanut rivers have temperatures ranging from 25-30℃ with a water discharge ranging from 25-40 L $second^{-1}$.

2.2 Ethical approval

All animal experimental and rearing procedures were handled complied with the animal welfare under Indonesian National Standard No. 6141:2009 about production of black tilapia fry *Oreochromis niloticus* Bleeker.

2.3 Fish Sampling

The fish that we sampled is tilapia fry stain red tilapia *Oreochromis niloticus* with size ±4-6 cm. Sampling fish were taken randomly at Working Unit of Fresh Water Fish Hatchery Loka Janti, Klaten. Sampling was carried out in 28 semi-permanent ponds with a total pond area varied between 133.65-952.00 m^2. The pond construction is equipped with a pair of inlet and outlet pipes measuring 6 inches and a pair of inlet and outlet sluice gates with a width of ± 40 cm. Each sample was taken as many as 5 samples in each 28 semi-permanent ponds. Observation of fish samples were put into sample bottles that had been given water and oxygen to be taken to the laboratory.

2.4 Fish Physical Examination

The physical examination of fish is designed to detect the presence of ectoparasite infestations on the gills, mucus, and fins. This involves a thorough inspection from head to tail, meticulously observing for any signs of ectoparasites. This detailed examination is essential for maintaining fish health, as untreated ectoparasite infestations can cause significant harm. Physical measurements were also carried out to obtain data on standard length, specific growth rate (SGR), and survival rate (SR) by measuring fish body length and weight. The first step of fish physical examination is measure the standard length and weigh the weight of tilapia fish fry. The second step of physical examination involves inspecting and made a smear preparation from each organ for ectoparasite infestations. This is done by collecting a mucus sample and making an incision to examine the gills and fins closely. This thorough process helps detect any signs of ectoparasites, ensuring a comprehesive assessment of the health of fish. The observation method is the same for both nursery 1 and nursery 2.

2.5 Ectoparasite Examination

The ectoparasites from fish collection from gills was started with incision on the ventral side of fish close to fish operculum, starting from the cloaca area to the anterior area of the fish, then take a pinch of gill filament with surgical scissors. Fish gill filaments were placed in a disposable petri dish containing 0.9% physiological NaCl. This organ is not stored in the refrigerator at 4°C, as it is observed directly under the Olympus CX23 binocular microscope using the 10x objective lens. Before observation, the gill filament is finely chopped for easier examination under the microscope. The chopped gill filaments are then placed on a glass slide and covered with a cover glass. Lugol's iodine solution is added to slow down the movement of ectoparasites, making it easier to observe their body shape and count their numbers.

2.6 Parameters

2.6.1 Water Quality

The observed water quality includes physical and chemical parameters that are measured during fish hatchery activities. Water samples were collected from the dept of

50-60 cm from the surface using the water quality checker Lutron DO-5510 Digital Oxygen Meter and Lutron pH 207. Water quality parameters measured each pond such as dissolved oxygen (DO), water temperature, and pH level. The dissolved oxygen (mg/L) was measured using a digital oxygen meter Lutron DO-5510 Digital Oxygen Meter which is equipped with a polarographic type probe with a temperature sensor that functions to measure DO in water, oxygen (O2) in the air as well as measure water temperature. Measurements of DO, water temperature and water pH are carried out directly in each pond. Water pH was measured using a pH meter Lutron pH 207.

2.6.2 Measurement amount of parasites

The number of each parasite species and the total number of all parasites in each pond was calculated to determine the Prevalence (P), Intensity (I), Mean Intensity (mI) and Mean Abundance (mA) by using the mathematical calculations formulated by (Bush et al. (1997). The prevalence (P) is the number of hosts infected with one or more individuals of a particular parasite species divided by the number of examined hosts. It describes the proportion of infected fish with one parasite species. The intensity (I) shows the range from the lowest number of parasites to the highest number of parasites in a fish of a certain species. The mean intensity (mI) is the average number of individuals of a parasite species, which can theoretically be found in an infected host. It is the total number of parasites in the sample divided by the number of hosts infected with that parasite. The main abundance (mA) is the number of individuals of a particular parasite in a single fish without taking into consideration whether the fish is infected or not. The following is a formula for calculating parasites:

$$P\ (\%) = \frac{Number\ of\ infected\ fish\ with\ one\ parasite\ species}{Number\ of\ examined\ fish}\ x\ 100$$

$$mI = \frac{Total\ number\ of\ one\ parasite\ species}{Number\ of\ fish\ infected\ by\ that\ species}$$

$$mA = \frac{Total\ number\ of\ one\ parasite\ species}{Number\ of\ examined\ fish\ by\ one\ species}$$

2.6.3 Fish growth and survival

The measurement of fish growth in fish hatchery was calculated with specific growth rate (SGR) and survival with survival rate (SR) formula for one cycle. These parameters measured from phase I and II nursery. Phase I nursery is the maintenance of 1-3 cm sized fry to 3-5 cm, while phase II nursery is the maintenance of 3-5 cm sized fry. Fish body weight measurement using digital balance with a precision of with ± 0.1 g. Fish growth and survival rates were calculated as described by by Nimrat *et al.* (2011) using the formula below:

$$Specific\ growth\ rate\ (SGR) = \frac{(lnW2 - lnW1)}{t2 - t1}\ x\ 100$$

where ln is the logarithmic number, w1 is initial weight at time t1 and w2 is the final weight at time t.

$$Survival\ rate\ (SR) = \frac{Fish\ number\ at\ the\ \ of\ experiment}{Fish\ number\ at\ the\ beginning\ of\ experiment}\ x\ 100$$

2.6.4 Financial value

Financial value measurement is carried out for 1 year of hatchery business activities where there are 10 cycles of fish hatchery. The calculation of financial value used calculating business costs which include investment costs, fixed costs, and variable costs. Furthermore, the calculated business analysis includes profit, R/C ratio, break event point (BEP), and payback period (PP). Profit is the value of the difference between revenue and the total cost of production. If the revenue is greater or has a positive value than the total cost of production, it is called profit. The value of the R/C ratio is the ratio between the total revenue and the total cost of production. R/C ratio is often used to analyse the feasibility of a business. A business is called to be profitable if the R/C ratio is more than 1.0, if it is equal to 1.0 it is called to be a breakeven point, and if it is less than 1.0 then it is called to be a loss. BEP is a condition where the level of sales of a business reaches the breakeven point, namely no profit and no loss. PP is the payback period for investment capital that has been issued at the beginning of business establishment. Cost of goods sold (COGS) is the cost of acquiring or manufacturing the products that a company sells during a period, so the only costs included in the measure are those that are directly tied to the production of the products, including the cost of labour, materials, and manufacturing overhead. The following is the formula used to calculate financial value.

$$\text{Profit} = \text{total revenue} - \text{total cost} \quad ; \quad \text{R/C ratio} = \text{total revenue/total cost}$$

$$\text{BEP (IDR)} = \frac{fixed\ cost}{1 - \frac{variable\ cost}{total\ revenue}} \quad ; \quad \text{BEP (Unit)} = \frac{fixed\ cost}{1 - \frac{variable\ cost}{production\ amount}}$$

$$\text{PP} = \frac{investment\ cost}{profit}\ x\ 1\ year \quad ; \quad \text{HPP} = \frac{total\ cost}{total\ production}$$

2.7 Data Analysis

The data of water quality parameters, SR, SGR, parasite counts and business analysis were tabulated in Microsoft Excel 2011. All data obtained were analyzed descriptively using tables and figures.

3. RESULTS

3.1 Water Quality

Measurements indicate that the water temperature (25.5–29.0 °C) falls within the range compliant with Indonesian National Standard No. 6141:2009. However, dissolved oxygen (DO) concentrations vary, with some results falling below the standard (>5 mg

L⁻¹), which may induce stress on fish. Additionally, the water pH (7.8–9.0) slightly exceeds the SNI limit (6.5–8.5). Overall, the environmental conditions in the pond require ongoing monitoring, particularly for DO and pH parameters, to maintain the stability of the aquatic ecosystem.

3.2 Measurement amount of parasites

The results obtained from the table above show that the highest prevalence (P) is 100% *Trichodina* sp., 60% *Dactylogyrus* sp., and 80% G*yrodactylus* sp. Intensity (I) and mean intensity (mI) respectively for *Trichodina* sp. are 48–211 and 93.6, *Dactylogyrus* sp. are 2–4 and 3.33, and for *Gyrodactylu*s sp. are 1–7 and 3.75. The mean abundance (MA) values of *Trichodina* sp., *Dactylogyrus* sp., and *Gyrodactylus* sp. respectively is 93.6; 2; and 3. The results of observing ectoparasites under a microscope can be seen in Figure 1.

3.3 Fish growth and survival

The data shows that the percentage of SR from the first nursery phase in Loka Janti is very low with an average of only 30.3%, while for the second nursery phase it has a higher value reaching 63.4%. The SGR value in nursery phase 1 with size of ± 4-6 cm was 15.6%.

3.4 Financial value

Based on the calculation results, it was found that there was a parasite attack on red tilapia fry with an SR of only 30.3%, but it still provided a profit of IDR 71,814,554.00/year. However, when viewed from the value of the payback period of 4.4 years, this means that this business will return capital up to 4.4 years.

4. DISCUSSION

The results obtained showed that the highest prevalence (P) 100%, Intensity (I) 48–211, and mean intensity (mI) 93.6, and mean abundance (mA) 93.6 is *Trichodina* sp. compared to other parasites in fish samples. In this study, many ectoparasites *Trichodina* sp. were found among other types of ectoparasites. This is because the environmental conditions of the rearing ponds and the accumulation of nutrients from feed residues trigger faster growth of ectoparasites (Purbomartono 2010). Ohoiulun (2002) reported that the surface of the body fish is directly related to the environment which facilitates the attack of ectoparasites including *Trichodina* sp. This type of ectoparasites is more common on the surface of the body fish than on other organs because it contains a lot of mucus and epithelial tissue which is a good place to live for ectoparasites and a place to find food. Parasites that attack these seeds are still categorized as safe and do not interfere with the production process.

Trichodina sp. has several variations in shape such as a bell as a sucker from chitin which resembles a circular anchor around the mouth (Gusrina 2008). *Trichodina* sp. has a role in reducing the immune system of fish, causing secondary infections (Rukmawa 2005). In Table 1, *Trichodina* sp. many attack the gills because it is in line with (Handayani 2020) that *Trichodina* sp. eat red blood cells which are found in the gills. *Trichodina* sp. uses the host only as a substrate and takes organic particles from bacteria, but the attachment of *Trichodina* sp. often cause injuries (Gusrina 2008). *Trichodina* sp. will easily grow if the water quality decreases, as a result the appetite of fish will be disturbed, and it will reduce the level of sensitivity to bacterial infections so that it can cause large losses. Meanwhile, a high infection rate can cause acute death without preceded by symptoms (Bhakti 2011).

Gyrodactylus sp. is an ectoparasite that attacks the skin and gills of freshwater fish. These ectoparasites are viviparous, where the eggs released will develop and hatch in the uterus (Noga 1996). *Gyrodactylus* sp. It infects the gills of the fish, the fish looks gasping for breath as if they lack oxygen, there is a lot of fish mucus production and the fish swims on the surface of the water. *Gyrodactylus* sp. has a small, elongated body. It has two ear-like protrusions at its anterior end. Posteriorly there is an ophisthaptor with 16 marginal hooks.

Dactylogyrus sp. Found in the fins and gills of fish which are important organs for respiration. *Dactylogyrus* sp. including low-level worm parasites (Trematodes). Live without an intermediate host, so that its whole life functions as a parasite. Characteristics of *Dactylogyrus* sp. marked by the presence of two pairs of eyes and four protrusions on the anterior (Safutra 2006). Kriswinarto (2002) reported that fish attacked by *Dactylogyrus* sp. usually it will become thin, swim jerkily, the gill covers cannot close completely because the gills are damaged, and the skin of fish looks no longer transparent. Based on the calculation results, it was found that there was a parasite attack on red tilapia seeds with an SR of only 30.3%, but it still provided a profit of IDR 71,814,554.00/year. However, when viewed from the value of the payback period of 4.4 years, this means that this business will return capital up to 4.4 years.

Trichodina sp. is a protozoan parasite that can result in extremely high mortality rates particularly in tilapia fry. It can destroy the skin and gill epithelium of the fish, as well as it leads to the secondary infections by other pathogens, such as bacteria and fungi, which further stress the host leading to mortality (Vallado *et al.* 2016). *Dactylogyrus* spp. is highly host-specific monogenean ectoparasites and commonly found embedded in the gill tissues of farmed cyprinid fish as well as major problems for aquaculture (Li *et al.*

2022). Its infection is often related to a series of infectious disease outbreaks in commercial farms causing several fish species to have significant morbidity rates (Thoney and Hargis, 1991; Kritsky and Heckmann 2002; Jaruboonyakorn P *et al.* 2022). Monogeneans in the genus *Gyrodactylus* tend to be highly pathogenic to fish and have become a challenge to the aquaculture industry. These parasites have the potential to endanger the survival of wild fish populations (Anshary *et al.* 2023).

5. CONCLUSION

The presence of parasites in fish can lead to decreased development and appetite, significantly impacting growth rates. This situation causes the investment in fish feed to be disproportionate to the growth of the fish, resulting in increased maintenance costs and lower profits, particularly in Loka Janti, Klaten, Central Java. To mitigate this issue, it is recommended to maximize production, implement pond calcification and fertilization, and enforce biosecurity measures to prevent disease transmission and improve overall fish health and profitability.

CONFLICT OF INTEREST

We certify that there is no conflict of interest with any financial, personal, or other relationships with other people or organization related to the material discussed in the manuscript.

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Tabel 1 Water quality data in nursery ponds I and II

Measured parameters	Results	Indonesian National Standard (INS)
Temperature	25,5–29 °C	23–30°C
Dissolved oxygen	3,2–6,0 mg L^{-1}	>5 mg L^{-1}
pH	7,8–9,0	6,5–8,5

Table 2 Data on the results of calculating ectoparasitic parameters on red tilapia seeds *Oreochromis niloticus*

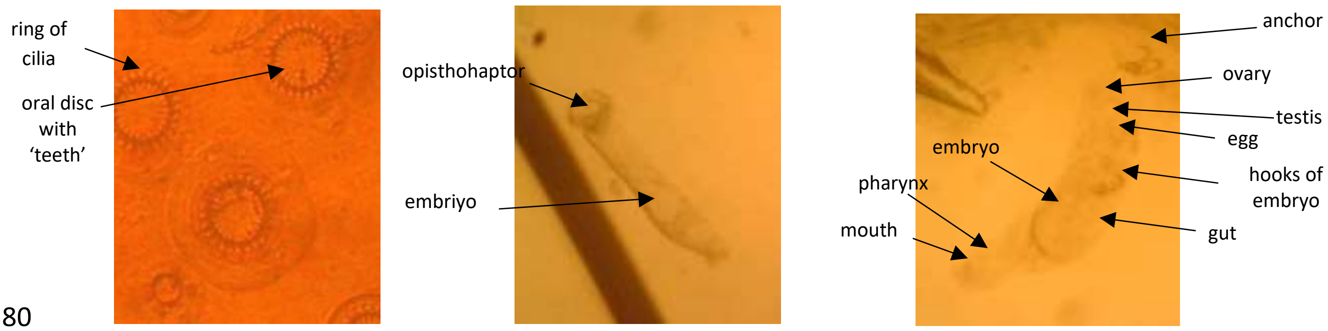
Ectoparasites	Parameter Value			
	P (%)	I	MI	MA
Trichodina sp.	100.00	48.00–211.00	93.60	93.60
Gyrodactylus sp.	80.00	1.00–7.00	3.75	3.00
Dactylogyrus sp.	60.00	2.00–4.00	3.33	2.00

Tabel 3 Survival rate and specific growth rate data for seeds in nursery phases I and II

Nursery Phase	Average Stocking (ind)	Average Harvesting (ind)	SR Average (%)	SGR (% day^{-1})
Nursery I	95.000	28.520	30,3	15.6
Nursery II	50.600	31.248	63,4%	2.2

Table 4. Analysis of red tilapia fish hatchery at nursery 1 size 4-6 cm

No	Cost and component calculation	Calculation results
1	Investment cost	Rp315,351,300.00
2	Cost of depreciation	Rp 21,775,133.00/year
3	Fixed cost	Rp256,199,146.00/year
4	Variable costs	Rp 98,049,000.00/year
5	Total cost	Rp354,248,146.00/year
6	SR	30.3%
7	Number of harvests 1 cycle	90,172 fry/cycle
8	Harvest cycle in 1 year	63 cycles
9	Total harvest in 1 year 63 cycles	5,680,836 fry/year
10	Selling price of fish	Rp75.00/fry
11	Total revenue (TR) in 1 year	Rp426,062,700.00/year
12	Profit	Rp 71,814,554.00/year
13	R/C ratio	1.20
14	BEP (Rp)	Rp332,781,527.00
15	BEP (Unit)	4,437,087 units
16	PP	4.4 years
17	HPP	Rp 62.00/fry



(a) (b) (c)

Figure 1. Ectoparasites found in red tilapia fry measuring 4-6 cm in Janti Loka: (a) *Trichodina* sp. (b) *Gyrodactylus* sp. (c) *Dactylogyrus* sp.