Techniques for bloodworm (Chironomid larvae: Diptera) mass culture in tarpaulin tanks

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Abstract. The aim of this study was to develop a culture technique that can be applied for mass production of bloodworms (Chironomidae larvae). Culture of bloodworms was carried out in outdoors place covered by a polycarbonate canopy. Culture of bloodworms used two culture units, first: spawning unit used to produce bloodworm eggs, and second: bloodworms rearing unit. Our study showed that the spawning unit could produce 322.67±9.71 egg masses in 3 days, and must be spread in eight rearing units for 12 days. Biomass of the bloodworms that were fed with fish manure, chicken manure and commercial fish feed were 194.8±7.3 g $tank^{-1}$, 150.8±9.8 g $tank^{-1}$ and 167.1±9.3 g $tank^{-1}$ respectively. Nutrition contents of bloodworms from this culture were 39.4-50.81% protein, 1.70-4.18% fat, and 22.94-25.76% carbohydrate. Fish manure and chicken manure are suitable for bloodworm feed because they are cheap.

Key Words: live feed, egg mass, chironomids, fish manure, chicken manure, ornamental fish feed.

Introduction. Currently, keeping ornamental fish is becoming increasingly popular for many people. Ornamental fish has turned into a big industry and has become one of the alternatives to fish farming besides consumption fish (Monticini 2010; Dey 2016). Live food has an important contribution to the development of ornamental fish culture (Lim et al 2003). Bloodworm is an insect larvae from the family Chironomidae known as non-biting fly which is a favorite live food for ornamental fish (Das et al 2012).

Bloodworm is considered as superior feed for ornamental fish because it has suitable nutrients for freshwater fishes (Thipkonglars et al 2010) with high protein content reaching 52.11-60.15% (Fard et al 2014; Ghazwan 2015). The average energy density contained in the bloodworm is 2.46 kJ g^{-1} (Meyer et al 2016). Bloodworm is a live food favored by fish (Gupta & Banerjee 2009; Anogwih & Makanjuola 2010; Baki et al 2015), providing optimal fish growth (Ghazwan 2015; Dey et al 2016; To'bungan 2016; Priya et al 2018), and increase fish pigmentation (Maleknejad et al 2014). The need for bloodworms continues to increase along with the increasing interest in keeping ornamental fish throughout the world. Unfortunately, most bloodworms used for ornamental fish feed are harvested from natural waters, and not from culture effort, so sometimes its quality is low and its availability is depending on season (Sulistiyarto & Christiana 2015; Nath et al 2017). Over exploitation of bloodworm in natural waters can disturb the ecological balance (Reynolds & Peres 2006). To meet the needs of bloodworms for the ornamental fish industry, mass production of bloodworms is needed.

The studies of bloodworm culture have been carried out using various feed sources including: cow dung, chicken manure, fish manure, vegetable waste, phytoplankton, yeast (Shaw & Mark 1980; Kumar & Ramesh 2014; Maleknejad et al 2014; Hamidoghli et al 2014; Suhada & Syamsudin 2014; Kumar 2016; Sulistiyarto & Restu 2018). Shaw & Mark (1980) cultured bloodworms in ponds so that the production was quite abundant, while other studies were carried out on a small scale in the Laboratory. The aim of our study is to develop a culture technique that can be applied for mass production of bloodworms.

Material and Method. This study was conducted at the Aquaculture Laboratory, Palangkaraya Christian University from May to August 2019. Culture of bloodworms was carried out in outdoors place covered by a polycarbonate canopy. The bloodworm broods were obtained from a culture collection maintained by the Aquaculture Laboratory of Palangka Raya Christian University. Culture of bloodworms used two culture units, first: spawning unit used to produce bloodworm eggs, and second: bloodworms rearing unit. Spawning unit used a plastic container with a size of 20 x 38 x 75 cm filled with 40 L of water. The top of the container was covered with a cage of mosquito net with a height of 50 cm. Rearing units used 9 (nine) tarpaulin tanks with a size of 100 x 100 cm, and filled with water to a depth of 20 cm (200 L).

Around 800-1000 individuals of 8-day-old of bloodworm broods were kept in spawning unit. The bloodworm broods were fed with commercial fish feed at the rate of 1.5 g L^{-1}. About 4 days later this spawning unit produces a lot of egg masses. The egg masses were then distributed in tarpaulin tanks. Each tank was spread with 40 egg masses. Three diets were studied including fish manure, chicken manure, and commercial fish feed. Fish manure and chicken manure were dried under sunlight for 3 days, ground and sieved, and then used as bloodworm feed. Feeds were offered at the rate of 1.5 g L^{-1}. Bloodworm was reared in tarpaulin tank until 12 day old. Bloodworm was harvested using scoop net with 1 mm mesh size and then rinsed under flowing water to wash away from dirt material. Survival rates, body length and biomass of bloodworms were measured. Water quality parameters of rearing tanks were observed including water temperature, pH, dissolved oxygen, and total dissolved solid (TDS). Nutrition contents of bloodworm (crude protein, crude fat, crude carbohydrate and crude fiber) were determined according to AOAC (2000). Data from this study were analyzed using one way ANOVA and followed by the LSD (Least Significant Difference) test.

Results. In this study, spawning unit produced 322.67±9.71 egg masses in one culture cycle. Each egg mass contained 296.22±82.05 eggs. For the next stage of culture, these egg masses require eight rearing tanks, because each rearing tank can only be spread with 40 egg masses. The survival rate of larvae during rearing in tarpaulin tanks using fish manure, chicken manure, and commercial fish feed as feed were 0.67±0.02%, 0.58±0.02% and 0.53±0.05% respectively.

Biomass and body length of bloodworms that were fed with fish manure, chicken manure and commercial fish feed are presented at Table 1. Results of analysis of variance (ANOVA) showed that different feeding regimes significantly influenced bloodworm biomass (F_{value} = 18.82). The LSD test indicated that the use of fish manure produced the highest bloodworm biomass, followed by commercial fish feed and chicken manure. The feeding regimes did not significantly effect on bloodworm body length (F_{value} = 0.33).

Table 1

The biomass and body length of bloodworm with different feeding regimes

Feeding regimes	*Biomass (g tank^{-1})*	*Body length (mm)*
Fish manure	194.8±7.3^{a}	9.610±0.058^{a}
Chicken manure	150.8±9.8^{b}	9.940±0.470^{a}
Commercial fish feed	167.1±9.3^{b}	9.892±0.800^{a}

The water quality conditions of the rearing tank are presented in Table 2. The use of different feeds affected water quality, especially TDS and pH. The use of fish manure produced high TDS and pH compared to the use of commercial fish feed and chicken manure.

Table 2

Water quality parameters of tarpaulin tanks

Parameters	*Feeding regimes*		
	Fish manure	*Chicken manure*	*Commercial fish feed*
Temperature (^oC)	27.68±0.24	27.67±0.19	27.55±0.24
TDS (mg L^{-1})	111.83±7.11	77.67±5.54	55.67±6.83
pH	7.73±0.02	6.28±0.05	7.13±0.09
Dissolved oxygen (mg L^{-1})	3.17±0.40	3.60±0.45	3.98±0.21

Bloodworms from this culture contained 39.40-50.81% protein, 1.70-4.18% fat, and 22.94-25.76% carbohydrate (Table 3). Results of analysis of variance showed that feeding regimes significantly affect on protein content (F_{value} = 84.66) and fat content (F_{value} = 158.12). LSD test indicated that highest protein content was obtained by use of chicken manure, meanwhile the use of commercial fish feed and fish manure were not significant different. The highest fat content was obtained by use of chicken manure and commercial fish feed and followed by fish manure. Carbohydrate content was not significant different between feeding regimes (F_{value} = 3.73).

Table 3

Nutrition contents of bloodworm with different feeding regimes

Feeding regimes	*Crude protein (%)*	*Crude fat (%)*	*Crude carbohydrate (%)*	*Crude fiber (%)*
Fish manure	39.40±0.96^b	1.70±0.14^b	24.80±0.98^a	5.66±0.07
Chicken manure	50.81±1.12^a	4.18±0.18^a	22.94±1.52^a	2.33±0.15
Commercial fish feed	43.59±1.17^b	3.42±0.21^a	25.76±1.29^a	3.36±0.16

Discussion. Culture of bloodworms requires the continuous availability of bloodworm eggs. The results of this study indicate that egg mass supply can be carried out sustainably using spawning unit. The spawning unit was able to produce about 322 eggs masses of bloodworm in one culture cycle which takes about one week or 1288 eggs masses for a month. Egg masses which were produced contain 296.22±82.05 eggs/egg mass. Egg content of bloodworm egg mass in this study is not much different compared to other studies, such as Kumar (2016) who obtained 286.66±1.24 eggs/egg mass and Kuvangkadilok (1994) who obtained 238.40-474.33 eggs/egg mass.

The survival rate of bloodworms during culture was relatively low when compared to another study, such as Podder et al (2018) states that the survival rate of bloodworms was 87.66-95.33%. To increase the survival rate of bloodworms requires better water quality control. The decaying process of bloodworm feed used, especially commercial fish feeds, contributes to water quality degradation.

The yield of bloodworm biomass from this culture ranged from 150.8 to 194.8 g $tank^{-1}$, depending on the feeding regimes used (Table 1). Bloodworm culture conducted by Shaw & Mark (1980) in ponds using chicken manure as food source produced 28 g m^{-2} biomass, while Shafruddin et al (2006) produced 88.44 g m^{-2} biomass. Thus the culture carried out in this study produced higher bloodworm biomass. In one culture cycle, about 322 egg masses of bloodworm were produced, and then grown in eight rearing tanks would produce about 1206.4-1558.4 g of bloodworm biomass.

The condition of the water temperature, pH, and DO of the rearing tanks were still within the range that could be tolerated by bloodworms. According to Baek et al (2012) for the development of bloodworm requires a water temperature of 25-30°C. Pinder (1986) states that pH 6-9 can be tolerated by bloodworms. Optimal dissolved oxygen levels for bloodworm culture are at least 3 mg L^{-1} (Lawrence 1981).

Protein is a very important component of fish nutrition because protein provides energy and growth performance of fish (Prabu et al 2017). Protein requirements in fish feed differ depending on the life stage, and food habits of fish species. The crude protein requirements in many ornamental fishes range between 25 to 55% (Velasco-Santamaría

& Corredor-Santamaría 2011). Bloodworms from this culture were able to meet the protein requirements for many ornamental fish species. The protein content of bloodworm from this culture was not too different from other studies. The protein content of bloodworms from other studies were 41.8% (Jayalekshmi et al 2017) and 55.4% (Davis & Raja 2019). The use of chicken manure as a food source produced bloodworms with higher protein content compared to using commercial pellets and fish manure. Chicken manure is a cheap and good feed material for mass production of bloodworms. Fat content in feed is an important source of energy (Prabu et al 2107), and fatty acids that are essential for normal growth and survival of fish (Sales & Janssens 2003). The fat content of bloodworms from this culture was quite low when compared with other studies. The fat content of bloodworms from other studies were 9.17% (Jayalekshmi et al 2017) and 8.12% (Davis & Raja 2019). Carbohydrate can be used limitedly for fish energy sources. According to Earle (1995) carbohydrate is not an essential nutrient for fish. The carbohydrate content of bloodworms from this culture was quite high (Table 3). The study conducted by Davis & Raja (2019) found that carbohydrate content of bloodworms was 18.01%. According to Gatlin (2010) carbohydrate content in feed for carnivorous fish is less than 20%, while for omnivorous fish contains 25-45%. Carbohydrate digestibility can vary from 70% in goldfish (*Carassius auratus*) to as low as 50% for moonlight gourami (*Trichogaster microlepis*) (Sales & Janssens 2003). The crude fiber content is not a nutritional component for fish and is not digested by fish. Based on Table 3, the crude fiber content of bloodworms from this culture ranges from 2.33 to 5.66%. The crude fiber content of bloodworms from this study was still suitable for fish feed. Jayalekshmi et al (2017) found that bloodworm contained 2.9% crude fiber. The crude fiber content suitable for fish feed is less than 7% (Gatlin 2010).

Conclusions. For mass production of bloodworms, we need two culture unit, that are spawning unit for produce bloodworm egg masses, and bloodworm rearing unit. Our study showed that the spawning unit could produce 322.67±9.71 egg masses in 3 days, and must be spread in eight rearing units of 1 x 1 m tarpaulin tanks for 12 days. This culture produced bloodworm biomass in range of 150.8-194.8 g tank^{-1} or 1206.4-1558.4 g/culture cycle. Nutrition content of bloodworms from culture conducted were 39.4-50.81% protein, 1.70-4.18% fat, and 22.94-25.76% carbohydrate. Food sources which can be used for bloodworm culture are fish manure, chicken manure, and commercial fish feed, but fish manure and chicken manure are more suitable because they are cheap source of feeds.

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