Feeding combination of live and fresh silkworms (*Tubifex* sp.) for the growth performance and survival rate of larvae of baung (*Hemibagrus nemurus*)

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Abstract. Economically important riverine fish species of baung (*Hemibagrus nemurus*) have been controllable spawned in a hatchery. The larvae rearing relies on expensive live feed of *Artemia* sp. Therefore, the experiment aimed to evaluate various feeding combinations of the live and fresh silk worms (*Tubifex* sp) in the rearing of the larvae of baung. The treatments were various combinations of live and fresh silkworms at 0%:100%, 25%:75%, 50 %:50%, 75%:25%, and 100%:0%. The seven-day-old larvae of baung of 0.0079 ± 0.019 g in weight and 8.96 ± 0.48 mm in length were randomly stocked into 15 plastic tanks of 40x25x17 cm in dimension size at a density of 160 larvae per tank and fed on a diet according to the treatment at 10%. of biomass per day for 21 days of the rearing period. The results showed that feeding silkworms in terms of live and fresh affected significantly the growth performance and survival of the larvae ($P<0.05$). The feeding combination at 75-100 % live and 25% fresh silk worms resulted in a high growth performance and survival of the baung larvae.

1 Introduction

One of the freshwater fish species having important economic value as table fish is baung (*Hemibagrus nemurus*). This species inhabits rivers in the Sunda Shelf, such as Indonesia (Java, Sumatra and Kalimantan), Malaysia and Thailand [1]. People living in the surrounding river of their habitat generally catch the baung fish seasonally and have traditionally cultured them relying on natural juveniles. Following time, the population of the being in nature tends to decline due to overfishing and a decrease in water quality. Fish culture is a must to meet the needs of the table fish and will reduce their fishing activities. The development of the baung culture requires a continuous juvenile supply from fish hatcheries [2].

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The performance of fish juvenile production requires the accuracy of initial feeding at the larval stage. Lack of feed and or feeding errors will lead to cannibalism of the baung larvae, which appear at the age of 54 hours [3]. The transition period to feed for African catfish larvae (*Clarias gariepinus*) occurs around 36-72 hours [4]. Furthermore, the increase in the survival rate of the larvae occurs through providing good environmental conditions, giving the appropriate first diet and avoiding delays in feeding.

Artemia sp. is the best natural feed for fish larvae, but the price is relatively high. Some efforts to reduce the use of *Artemia* sp. are shortening the feeding period and or looking for other species of natural feed as a substitute for *Artemia*. The baung larvae rely totally on *Artemia* sp. as the initial diet [5] and African catfish (*Clarias gariepinus*) [6]. The snakehead (*Channa striata*) larvae cultured in green media can accept *Daphnia* sp. as the initial diet [7]. *Daphnia* sp. enriched using a commercial diet increases the survival rate of larvae of torsoro (*Tor soro*) [8] and enriched by glutamine for survival larva of giant gouramy (*Osphronemus gouramy*) [9]. After the initial feeding is complete, the next feed used for the baung larvae is silkworms (*Tubifex* sp.) until the larvae can accept an artificial diet [10,11].

After going through the cleaning process, the next process is a reduction in the size of silkworms according to the size of the mouth of the larvae. But, the use of live silkworms yielded from nature have some weakness, namely, the carrier of microorganisms as a source of disease, limited availability, and relatively long storage can cause death and/or a decrease in quality. Providing fresh silkworms is an effort to reduce the use of live silkworms. Therefore, an experiment was conducted to evaluate the feeding of live and fresh silkworms on the growth performance and survival of seven old baung larvae.

2 Experimental Method

A completely randomized design with five treatments and three replications was used in this experiment. The treatments were various compositions of live and fresh silkworms. namely: 0%:100%; 25% :75%; 50% : 50%; 75% : 25%; 100%: 0%.

The test diet used was silkworms (*Tubifex* sp.) yielded from nature. The silkworms were treated as the following: washed thoroughly and separated from mud and dirt, soaked in one litre of water that had been given one drop of methylene blue for two minutes, and then placed in a container having running water and aerated sufficiently using an aerator. Some of the silkworms were taken and stored in a container in a freezer at a temperature below 0^oC. Before being given, frozen silkworms are left at room temperature until they thaw. Live and fresh silkworms were sampled as subjects for proximate analysis in an accredited laboratory (Table 1). When fed, frozen silkworms are thawed at room temperature. Silkworms were weighed daily every morning according to a specified amount of diet.

Table 1. Proximate composition of live silkworms and fresh (% dry weight)

Nutrients (%)	live silkworms	Fresh silkworms
Moisture	90.69	82.46
Crude protein	59.82	61.66
Crude lipid	11.29	10.66
Crude ash	3.58	5.24
Crude fibre	2.22	2.04
Nitrogen free extract	23.10	20.32

The test fish used were baung larvae collected from controllable spawning of a single population of brooder of the fish. On the second day, the larvae were fed on *Artemia* sp. for two days and continued with live silkworms for two days as a period of adaptation for the larvae to test diet and environmental conditions. The size of the larvae was 0.0079 ± 0.0190 g in weight and 8.96 ± 0.48 mm in length.

The seven-day-old larvae were randomly stocked into 15 plastic tanks of 40x25x17 cm3 in a dimension filled with a volume of 16 L of water at a density of 160 individuals per tank or 10 individuals. L^{-1} [12] and fed according to the treatment of 10% of body weight per day for 21 days of the rearing period. The daily test diet was carried out 5 times at 08.00, 11.00, 14.00, 17.00, and 20.00 WIB. Before the first feeding, the waste in each tank was taken out through siphoning, and the wasted water was replaced. Water temperature and pH were measured thrice daily in the morning, noon, and evening. A daily diet and fish death of the larvae was noted. The number 20 larvae were sampled from each tank as subjects to measure weight and total length. At harvest, the number of larvae in each tank was counted, taking a sample of as many as 20 larvae to measure individual total length and weight.

Test parameters include absolute weight gain (ΔW), absolute length gain (ΔL), feed conversion rate (FCR) and survival rate (SR) are estimated using a formula referring to [13], specific weight growth rate (SWGR) and specific length growth rate (SLGR) were calculated using the formula according to [14]. Feed intake (FI) was determined based on the daily amount of feed for 21 days of the rearing period.

$$\Delta W\ (g) = W_t - W_o \quad (1)$$
$$\Delta L\ (mm) = L_t - L_o \quad .(2)$$
$$SWGR\ (\%.day^{-1}) = 100\ x\ ((Ln\ W_t - Ln\ W_o) / 21days) \quad (3)$$
$$SLGR\ (\%.day^{-1}) = 100\ x\ ((Ln\ L_t - Ln\ L_o) / 21days) \quad (4)$$
$$FCR = FI / ((W_t + W_d) - W_o) \quad (5)$$
$$SR\ (\%) = 100\ x\ (N_t / N_o) \quad (6)$$

Where W_t is the weight of baung larvae at harvest, W_o is the weight of baung larvae at stock, L_t is the total length of baung larvae at 21 days, L_o is the total length of baung larvae at stock, W_d is the weight of dead baung larvae during the rearing period, N_t is the total baung larvae at yield, and N_o is the total baung larvae at stock.

The effect of the treatment on the parameters test used Analysis of Variance (ANOVA) at a 95% confidence interval. Duncan's test was used if the ANOVA results showed a significant difference. The distribution of the total length at each observation as well as the temperature and pH of the water, are explained descriptively.

3 Result and Discussion

3.1 Growth performance

Table 2 contains the growth performance of baung larvae fed various combinations of live and fresh silk worms for 20 days of the rearing period. The baung larvae prefer live silkworms at harvest time, as indicated by the weight and total length growth. The fewer fresh silkworms are given, the more baung larvae growth performance tends to increase. The administration of live silkworms can increase the growth of various fish larvae, including snakehead (*Channa striata*) [15], catfish (*Clarias* sp.) [16], goldfish (*Carrasius auratus*) [17] and angelfish (*Pterophyllum scalare*) [18].

The growth of the weight of baung larvae given various compositions of live and fresh silkworms for 21 days follows the exponential equation (**Fig. 1**), namely:

$$y\text{-}_{T1\ (0:100\%)} = 0.0008x^2 - 0.0061x + 0.0136;\ R^2 = 0.8617 \quad (7)$$
$$y\text{-}_{T2(25:75\%)} = 0.0005x^2 + 0.0004x + 0.0079;\ R^2 = 0.9947 \quad (8)$$
$$y\text{-}_{T3(50:50\%)} = 0.0005x^2 + 0.004x + 0.0061;\ R^2 = 0.9907 \quad (9)$$
$$y\text{-}_{T4(75:25\%)} = 0.0005x^2 + 0.0028x + 0.0058;\ R^2 = 0.9848 \quad (10)$$
$$y\text{-}_{T5(100:0\%)} = 0.0008x^2 + 0.001x + 0.0086;\ R^2 = 0.9777 \quad (11)$$

Table 2. Growth performance of baung larvae on various combinations of live and fresh silkworms

Parameters**	Live and fresh silkworm combination (%)*				
	0:100	25:75	50:50	75:25	100:0
FI (g)	146.45±7.85[a]	159.47±4.74[b]	157.45±0.52[b]	159.60±0.52[b]	160.87±4.88[b]
ΔW (g)	0.1839±0.0488[a]	0.2473±0.0159[b]	0.2555±0.0130[b]	0.3077±0.0233[bc]	0.3782±0.0508[c]
ΔL (mm)	3.79±0.70[a]	5.87±0.30[b]	7.24±0.17[c]	7.20±0.32[c]	8.36±0.11[d]
SWGR (%day^{-1})	16.74±1.77[a]	17.37±0.31[a]	17.82±0.52[ab]	18.43±0.38[ab]	19.42±0.64[b]
SLGR (%day^{-1})	1.76±0.28[a]	2.52±0.10[b]	2.96±0.05[c]	2.95±0.10[c]	3.06±0.42[c]
FCR	2.20±0.48[b]	1.55±0.34[a]	1.18±0.09[a]	1.13±0.04[a]	1.09±0.04[a]

* mean ± SD value in the same row with similar superscript are not significantly difference
** W_t = body weight of baung larvae at 21 days, L_t = body length of baung larvae at 21 days, FI = feed intake, ΔW = absolute weight gain, ΔL = absolute total length gain, SWGR = specific weight growth rate, SLGR = specific length growth rate, FCR = feed conversion rate

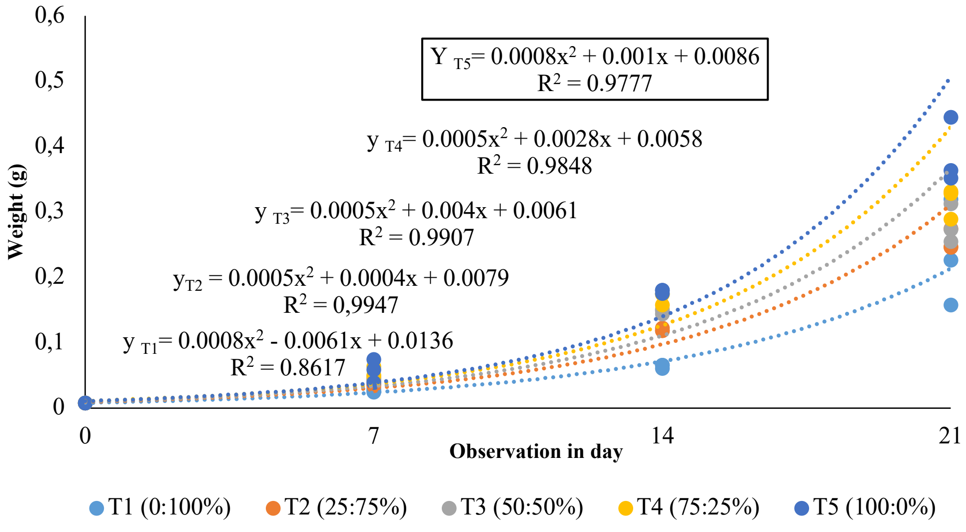


Fig. 1. Growth of baung larvae in various combinations of live and fresh silkworms during the 21-day rearing period

Fig. 1 shows that baung larvae tend to have adapted to silkworms in live and fresh started from day 7 of rearing. The exponential growth increase pattern proves that silkworms' nutrient content matches the nutritional needs of baung larvae (**Table 1**). However, the presence of live silkworms contributes to the appetite of baung larvae. This relates to silkworms being a natural feed that moves not very actively [17], so baung larvae easily prey on them. [10] added that baung larvae are carnivorous and like live natural feed in colonies such as silkworms.

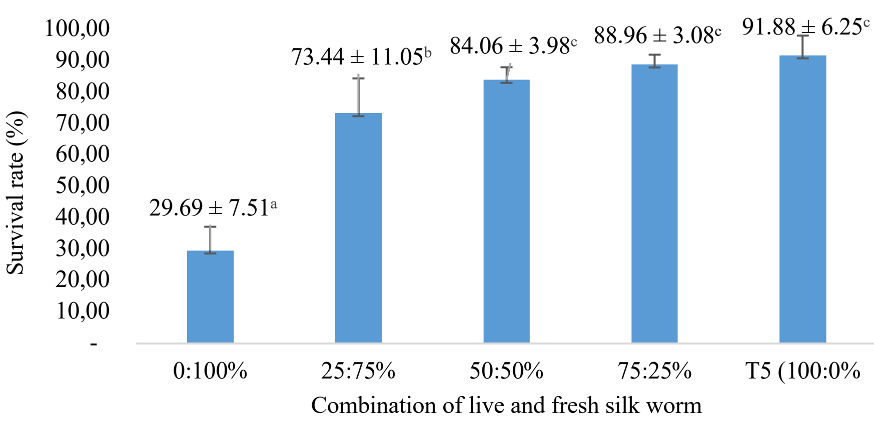
The performance of the growth of baung larvae was indicated by the value of the level of feed intake for each treatment, and the administration of 100% fresh silkworms resulted in the lowest feed intake ($P<0.05$). The administration of live silkworms ranging from 25 to 100% gave an increasing value of feed consumption and was no difference ($P>0.05$). Live silkworms have an attraction for the baung larvae to grasp them. Silkworms have high attractants to increase feed consumption in larvae [19]. According to [20], the smell and colour of live natural feed will be detected by chemosensory sensors so that the larvae can find and prey on it. Storage of silkworms in the refrigerator at 0°C is thought to contain low attractants, thereby reducing the appetite of the baung larvae.

The higher use of live silkworms led to increase absolute weight gain, absolute length gain and growth rate of specific length (P<0.05). This result is associated with increased feed intake to meet the nutritional needs of the baung larvae. Live silkworms contain high macro and micronutrients that contribute to the growth of larvae [21,22]. However, the results showed that the administration of 0-25% live silkworms resulted in a significantly difference specific weight growth rate from 100% live silkworms (P<0.05). The high presence of fresh silkworms is thought to result in the low growth of baung larvae. Live silkworms are able to have autolytic and contain exogenous enzymes that stimulate the formation of digestive enzymes, making it easier for the larvae to digest their food [23,24]. According to [25], frozen live natural feed causes low levels of exogenous enzymes, thereby reducing the activity of larval digestive enzymes [26] and growth [27]. Thus, the administration of low levels of fresh silkworms will increase the activity of larval digestive enzymes due to the high content of exogenous enzymes in live silkworms.

Feed quality can be reflected in the conversion value of feed into fish body meat. The smaller the feed conversion, the more feed is converted to meat. The results of this study showed that the highest feed conversion was achieved at 0% live silkworms (2.20 ± 0.48) and the lowest at 25-100% live silkworms (1.09-1.55) (P<0.05). The low feed conversion value in this treatment is due to the more efficient utilization of nutrients, and most of these nutrients were stored in the body of the baung larvae. Similar results are also reported [19] that the administration of 100% silkworms results in higher utilization of nutrients in snakehead (*Channa stroata*) larvae. The feed conversion values obtained were not much different from fish larvae in general that are fed on natural diet, both *Artemia* sp. [28], *Daphnia* sp. [29] and mosquito larvae [26].

3.2 Survival Rate

Fish deaths are generally caused by drastic changes in water quality, food shortages caused by fish's reduced appetite, and cannibalism due to lack of food. In this study, the temperature and pH of the water were in the medium for rearing baung larvae ranging from 25 - 30°C and pH 6.5 -7.0, respectively. This value is still within the normal range for the rearing media of baung larvae [5,10].



* mean ± SD value in the same row with similar superscript are not significantly difference

Fig. 2. Survival of baung larvae given various combinations of live and fresh silkworms during the 21-day rearing period.

The observations during the study showed that the death of baung larvae was not caused by cannibalism because no damaged body of baung larvae was found. The survival rate of baung larvae was significantly lower (29.69%) at 0% live silkworms, then significantly increased at 25% (73.44%) and 50-100% (84.06-91.88%) respectively ($P<0.05$). The more live silkworms contribute, the more baung larvae survive (**Fig. 2**). Proper feeding on natural feed is a determining factor of larval survival rate [30,31]. The existence of live silkworms will encourage baung larvae to grasp them so that the baung larvae will receive more nutrients and energy needed by these larvae. The results of this study prove that live silkworms contribute to the development, growth and survival of baung larvae.

4 Conclusion

The combination of live silkworms and 25% fresh silkworms at respective rate 75-100% and 25% resulted in the best growth performance and survival of baung larvae.

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