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Response of post-larva of snakehead (*Channa striata*) to feeding dried silkworm (*Tubifex* sp.) and artificial diet

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ABSTRACT

Post-larval snakehead (*Channa striata*) can be fed on live silkworms (*Tubifex* sp.), artificial feed and a combination of both as a source of nutrients. The availability of live silkworms depends on the yield in nature. Meanwhile, dried silkworms are available in the market. Therefore, the study to evaluate feeding combinations of dried silkworms and artificial feed for the rearing of post-larvae snakehead was conducted from October to December 2020 in Serang District, Indonesia. The treatments were combinations of dried silkworm and artificial feed at a ratio of 100:0%, 75:25%, 50:50%, 25:75%, and 0:100%, respectively. After being adapted to experimental conditions, post-larvae of snakehead (0.40±0.02 g) were randomly stocked in 15 happas of 50x70x75 cm in dimension size each at a rate of 25 post-larvae and fed on tested diet according to the treatments at 5% of biomass a day, divided into three times for 40 days of experimental periods. The post-larvae of snakehead performed the best growth at the contribution of dried silkworm at 100 – 25%, the best feed conversion and protein efficiency ratio at 100 – 50% and 50%, respectively and the best survival at 50 – 0%. It could be suggested that feeding dried silkworm and artificial feed at a 50:50 ratio, respectively, during post-larvae snakehead rearing.

Introduction

Snakehead (*Channa striata*) having an important economic value distributes in inland waters of Indonesia, such as Sumatra, Java, and Kalimantan (Hidayatullah *et al*., 2015). This fish also contains albumin as a biomedical material (Asikin and Kusumaningrum, 2018; Khasani and Astuti, 2019). This increases the snakehead demand, mostly fulfilled by catches in nature and threatens its sustainability. Many snakeheads were caught at the peak of the dry season and generally had gonads in a mature condition (Bijaksana, 2012). Domestication is the first step in preventing the extinction of snakeheads in nature and developing their culture.

Snakehead can ripen its gonad and spawn using hormonal induction (Muslim, 2019). The highest mortality of snakeheads occurs during larval rearing and this can be overcome through feeding management (Saputra *et al*., 2021). When the yolk is almost depleted, fish larvae need nutrients from their environment. This transitional period of nutrient sources becomes a critical point for the fish larvae rearing and will be an obstacle in developing snakehead culture (Janakiraman and Altaff, 2014; Paray *et al*., 2016). Lack of nutrients needed causes an increase in cannibalism of snakehead larvae (Paray *et al*., 2015). The nutrient source is live feed containing high protein, including *Daphnia* sp., proven to be able to substitute *Artemia* sp. on rearing snakehead larvae (Saputra *et al*., 2018), mahseer (*Tor douronensis*) (Ningsih *et al*., 2020), and gourami (*Osphronemus goramy*) (Fahmi *et al*., 2019). The next feed is live silkworms (*Tubifex* sp.) (Ndobe *et al*., 2017; Rahman *et al*., 2018).

Silkworms are among the best live feeds for larval rearing, both fish and shrimp, because they have high

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nutritional content, comparable with *Artemia* sp., and higher than *Daphnia magna* (Oplinger *et al.*, 2011; Herawati *et al.*, 2016). Live silkworms have no skeleton, so they are easily digested by fish larvae (Meilisza *et al.*, 2020). Feeding live silkworms on snakehead larvae can increase their growth and survival (Mahardika *et al.*, 2017). Live silkworms are also recommended for rearing larvae to fry for giant catfish (*Clarias gariepinus*) (Nuswantoro and Rahardjo, 2018) and catfish (*Pangasionodon* sp.) (Suprayudi *et al.*, 2013). However, the use of live feed, including silkworms, has disadvantages, namely that it cannot be stored for a relatively long time, its availability is limited in nature, and it is suspected to carry various diseases. Dried silkworms that have been widely used for ornamental fish are thought to be used for post-larvae of snakehead.

Post-larvae of fish can receive a nutrient from outside because their digestive organs and physiological functions are fully formed. Appropriate management and artificial feed are needed to support the successful domestication of snakehead fish (Nen *et al.*, 2018). Several methods to reduce live feeds are using live feed in combination with artificial feeds (Yulintine *et al.*, 2012). This has been done on the post-larvae of jelawat (*Leptobarbus hoeveni*) (Sunarno and Syamsunarno, 2017) and climbing perch (*Anabas testudenus*) (Rahmi *et al.*, 2016). Therefore, this study aimed to evaluate the appropriate combination of dried silkworms and artificial feed in the rearing of post-larvae snakehead.

Materials and Methods

Location and time of research

This research was conducted from October to December 2020 in fish farmer pond located in Baros, Serang District, Indonesia. A completely randomized design with five treatments and three replications was used in this study. The treatments were various combinations of dried silkworms and artificial feed at the following ratios of 100:0%, 75:25%, 50:50%, 25:75%, and 0:100%, respectively.

Dried silkworms and artificial feed meals were obtained from the market. Dried silkworms were floured and stored in labeled plastic bags. The two test feeds were sampled as subjects for proximate analysis at the Accredited Chemical Laboratory of the Research Institute for Freshwater Aquaculture and Fisheries Extension, Bogor. (Table 1). The proximate analysis method refers to Association of Official Analytical Chemists (2012).

The test feed was served daily to the test fish in the form of pasta. The silkworm and artificial feed were weighed according to the treatment and need, then put in a petri dish, mixed evenly, added enough water and mixed evenly to form a paste.

Table 1. Proximate composition of dried silkworm (*Tubifex* sp.) and artificial feed.

Tested diet	Composition (% in dry weight)				
	Crude protein	Crude fat	Ash	Crude fiber	NFE*
Artificial feed	41.12	6.54	0.82	1.85	39.67
Dried silkworm	61.13	6.21	0.89	2.11	25.66

Remark: *NFE= Nitrogen free extract

Experimental happa (tank)

Two unit happas having 2x1x1m in dimension size and placed in a $200m^2$ pond were used as acclimation tanks of snakehead post-larvae. The top part of the happa was closed by the net. The submerged part of the happa was about 50 cm. The water surface in the happa was covered with water hyacinth (*Eichhornia crassipes*) of about 25-50% of the total surficial area. For experimental tanks, 15 happas of 50x70x75cm in dimension size each were used.

Experimental fish

Larvae of snakehead were obtained from nature by catching them with a scoop simultaneously and from the same location. The yolk sac of the larvae was no longer visible. A total of 2000 snakehead larvae were reared in acclimatizing tank at a density of 1000 fish per tank and fed on test feed with a ratio of 50:50% at ad libitum in the morning, afternoon, and evening. The test feed was put in 4 discs placed in a storage happa with a depth of about 15 cm from the bottom and fed on the larvae for 15 days of the adaptation period. After that, the post-larvae of snakehead fish were yielded and selected to obtain a relatively uniform size and kept in a happa. The average weight of the post-larvae was 0.40±0.02 g.

The post-larvae rearing

The post-larvae of snakehead were weighed and then randomly stocked into the experimental tanks and repeated several times to fulfill the number of 25 fish per happa. After stocked, the post-larvae were not fed on test feed for 24 hours, after that, were given test feed according to the treatment as much as 5% of the total weight of the fish per day in the morning, afternoon, and evening for 40 days of fish rearing. The amount of test feed given was recorded every day. The test fish from each happa was counted and weighed by population every 10 days. The individual average weight was calculated by dividing the population weight by the number of fish that lived in each observation. Fish mortality was observed every day and weighed and observed in the laboratory.

Parameters

The parameters used in this study were absolute weight growth (ΔW), specific growth rate (SGR), feed intake (FI), feed conversion ratio (FCR), and survival rate (SR) which were calculated based on Handayani and Widodo (2010) and protein efficiency ratio (PER) calculated based on Nates (2016). The formula for calculating each parameter was as follows:

$$\Delta W\ (g) = Wt - Wo$$

$$SGR\ (\%/day) = \left[\sqrt[t]{\frac{Wt}{Wo}} - 1\right] x100\%$$

$$FCR = \frac{FC}{(Bt + Bd) - Bo}$$

$$PER\ (\%) = \frac{Wt - Wo}{Pi} x100\%$$

$$SR\ (\%) = \frac{\Sigma\ \text{fish larvae post at the harvest}}{\Sigma\ \text{fish larvae post at stocking}}\ x\ 100\%$$

Notes: Wt = weight at the end of rearing (g); Wo = weight at stocking (g); t= length of rearing (days); Bd= dead larval biomass during rearing (g); Bt= biomass at the end of rearing (g); Bo= biomass at stocking (g); Pi = feed protein consumed (g).

Data analysis

The data for each test parameter was tabulated and calculated using Microsoft Excel. The effect of treatment on test parameters was analyzed statistically by ANOVA at 95% confidence level. If the analysis results were significantly different, the test continued with Tukey's Test. Data was presented in the form of a picture.

Results

Figure 1 showed feed intake for post-larvae snakehead during 40 days of the rearing period. The use of dried silkworms in test feed ranging from 100% to 25% did not significantly affect the feed intake of post-larvae snakehead (P>0.05) and was significantly higher than that of 100% artificial feed (P<0.05). The best feed intake for snakehead post-larvae in this study ranged from 37.02 – 43.14 g.

The absolute weight growth of the post-larvae snakehead can be seen in Figure 2. The absolute weight growth was not significantly different at 100% to 25% dried silkworms in the test feed, ranging from 2.52 to 3.10 g (P>0.05). The lowest absolute weight growth value at 100% artificial feed was observed (P<0.05). The growth pattern of the specific weight of snakehead fish post-larvae and absolute weight growth was comparable (Figure 3). The best specific weight growth value was at 100% to 25% dry silkworms, ranging from 5.08% day^{-1} to 5.48% day^{-1} (P>0.05). The lowest specific weight growth value (P<0.05) in 100% artificial feed treatment (3.89±0.11% day^{-1}).

The protein efficiency ratio (PER) shows the total weight of fish produced from each unit of protein in the feed. The PER was highest at 100%-50% of dried silkworms in test feed and then decreased significantly at 25%-75% and 0%, respectively (P<0.05) (Figure 4).

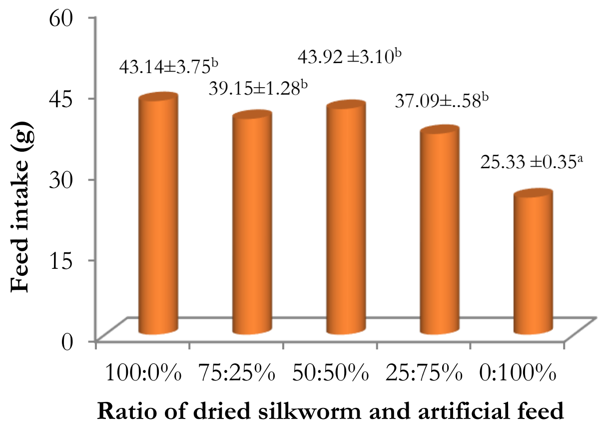

Figure 1. Feed intake of post-larvae snakehead (*C. striata*) for 40 days of the experimental period, where number behind ± indicates standard deviation value, similar letter in a similar row indicates no significant difference (P>0.05).

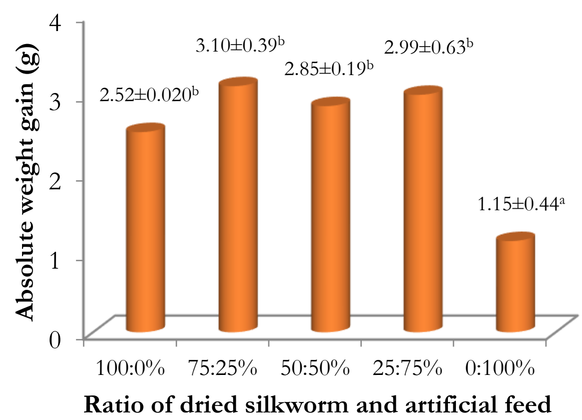

Figure 2. Absolute growth of snakehead (*C. striata*) post-larvae for 40 days of experimental period, where number behind ± indicates standard deviation value, similar letter in a similar row indicates no significant difference (P>0.05).

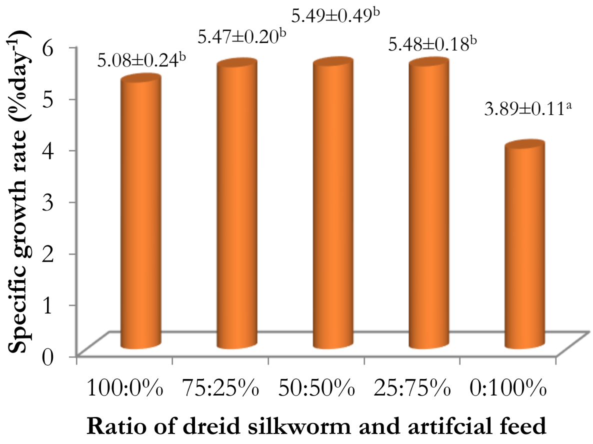

Figure 3. Specific growth rate of snakehead (*C. striata*) post-larvae for 40 days of the experimental period, where number behind ± indicates standard deviation value, similar letter in a similar row indicates no significant difference (P>0.05).

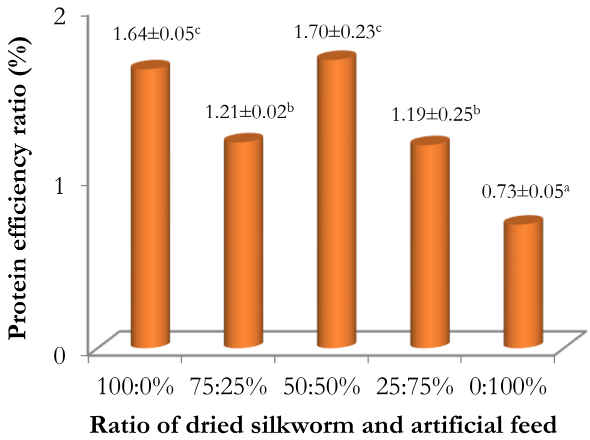

Figure 4. Protein efficiency ratio of snakehead (*C. striata*) post-larvae at the end of the rearing, where number behind ± indicates standard deviation value, similar letter in a similar row indicates no significant difference (P>0.05).

Figure 5 shows the feed conversion ratio (FCR) value for post-larvae snakeheads. The FCR value was influenced by various dried silkworms and artificial feed (P<0.05). The FCR values of snakehead post-larvae were not significantly different at 100% to 50% of dried silkworms (P>0.05) and were lower than that of 25% and 0% of dried silkworms (P<0.05). The best FCR ranges from 1.02-1.47. The worst FCR value (3.35) was at 100% artificial feed.

The survival rate (SR) of snakehead post-larvae on various combinations of dried silkworms and artificial feed can be seen in Figure 6. The Significant lowest SR value (44.00±5.66%) was observed at dried silkworms of 25% compared to other treatments (P<0.05). The SR of snakehead post-larvae at 100%, 50-0% of dried silkworms were not significantly different (P>0.05), ranging from 68.0% to 74.0%.

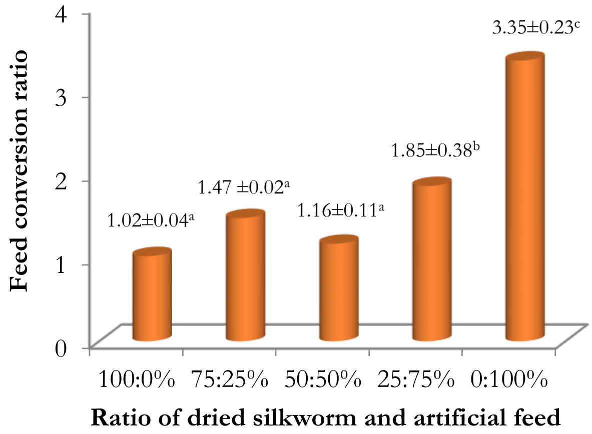

Figure 5. Feed conversion ratio of snakehead (*C. striata*) post-larvae at the end of culture, where number behind ± indicates standard deviation value, similar letter in a similar row indicates no significant difference (P>0.05).

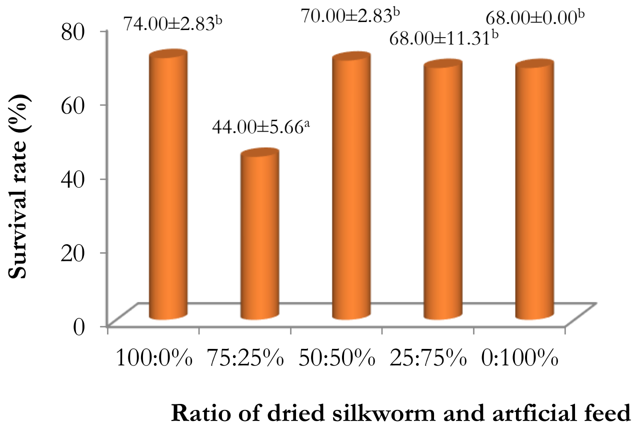

Figure 6. Survival rate of snakehead (*C. striata*) post-larvae at the end of rearing, number behind ± indicates standard deviation value, similar letter in a similar row indicates no significant difference (P>0.05).

Discussion

The use of dried silkworms in feed ranging from 100% to 25% did not affect feed intake of snakehead post-larvae (P>0.05) and was significantly higher than that of giving 100% artificial feed (P<0.05). It is suspected that dried silkworms have an attractant that stimulates the post-larvae of snakehead to consume them. The use of live feed in combination with artificial feed provides visual and chemical stimuli to increase the consumption of the post-larvae (Kolkovski *et al*., 1997a). The natural feed provides visual and chemical stimulation to increase feed consumption in fish larvae (Savona *et al*., 2011). The common attractant in 100% artificial feed is thought to decrease feed consumption of snakehead post-larvae. This is in line with the research results of Kolkovski *et al.* (1997b) that consumption of 100% artificial feed on gilthead sea bream (*Sparus aurata*) larvae was lower when compared to the combination of artificial feed and live feed.

Giving 100% artificial feed resulted in the lowest growth of snakehead post-larvae significantly than other treatments (Figures 2 and 3). This is associated with the lower nutrient content of artificial feed than dried silkworms (Table 1), so that the nutritional requirements for the growth of snakehead post-larvae are low. However, the use of dried silkworms combined with artificial feed fulfilled dietary needs so that growth in the treatment was higher and not significantly different ($P>0.05$). In addition, the low growth in the provision of 100% artificial feed is thought to be due to the incomplete digestive tract of snakehead post-larvae so that the absorption of nutrients in the stomach takes longer. Mullah *et al.* (2019) reported that artificial feeding resulted in longer gastric emptying time and decreased digestion rate, which correlated with lower metabolism and growth of fish larvae. Furthermore, Abol-Munafi *et al.* (2004) noted that snakehead post-larvae are could not digest artificial feed due to an imperfect digestive system and their nature as carnivorous fish.

The digestion rate of fish larvae is influenced by the nutritional composition of the feed and the activity of digestive enzymes. The natural feed has exogenous enzymes that can stimulate the production of endogenous enzymes in the digestive tract of fish (Nurhayati *et al*., 2014). Fish larvae can utilize enzymes containing in live feed for about 40-80% in the process of feed digestion (Savona *et al*., 2011). The enzyme is thought to still be present in dried silkworms so that it helps post-larvae of snakehead in digesting feed so that the utilization of nutrients, especially protein, is more efficient for growth. Based on Figure 4, the protein efficiency ratio of snakehead post-larvae on the administration of 100% dried silkworms was not significantly different from the provision of 50% dried silkworms and 50% artificial feed ($P> 0.05$). According to Chepkirui-Boit *et al.* (2011), a combination of live feed and artificial feed at 50:50%, respectively led to more optimal utilization of nutrients in fish larvae. This is associated with the content of exogenous enzymes that can help the digestive process of natural and artificial feeds be more efficient to increase protein utilization more optimally. Jusadi *et al.* (2015) added that the high activity of protease enzymes in the combination of silkworms and artificial feed resulted in optimal growth of fish larvae. The increasing use of artificial feed causes the lower protein efficiency of snakehead post-larvae. Fish larvae that consume artificial feed have low endogenous enzyme activity so that the utilization of nutrients is not optimal for growth (Kamarudin *et al*., 2011).

Efficient utilization of nutrients by snakehead post-larvae affects the value of feed conversion at the end of rearing (Figure 5). The use of artificial feed up to 50% in the test feed did not affect the feed conversion of snakehead post-larvae, with a value of 1.02-1.47 ($P>0.05$). A low feed conversion value indicates that more nutrients need for growth than for body maintenance. The combination of silkworms and artificial feed up to 50% can complement each other's advantages and disadvantages, resulting in a balance of nutrients needed by the snakehead post-larvae. The high value of feed conversion occurs along with the increase in the composition of artificial feed in the feed. This is thought to be due to the decreasing composition of dried silkworms so that digestive enzymes cannot help optimally digest artificial feed. The presence of silkworms in the feed results in a better digestive process so that artificial feed is more digestible due to the help of digestive enzymes in fish larvae (Rahmi *et al*., 2016). Economically, the high value of feed conversion will affect the cost of feed and affects production costs.

Post larval survival of snakehead fed a combination of dry silkworm and artificial feed for 40 days of rearing period ranged from 44-74% (Figure 6). The range of values is similar to the results of studies on the survival rate of climbing perch (*Anabas testudineus*) fed with various combinations of silkworms and artificial feed (Rahmi *et al*., 2016). Feeding combination of 75% dried silkworms and 25% artificial feed resulted in the lowest survival rate of snakehead post-larvae and was significantly different from other treatments ($P<0.05$). The low survival rate in this treatment is thought to be due to cannibalism. Based on daily observations that show no fish carcasses found in the happa. Cannibalism is one of the biggest problems in cultivating snakehead larvae which causes low survival (War and Altaff, 2014). The occurrence of cannibalism can be associated with fish larvae size that is not uniform and lack of feed in larval rearing (Abol-Munafi *et al*., 2004). However, the results showed that giving up to 100% artificial feed could increase the survival of snakehead post-larval and were not significantly different from giving 100% dried silkworms. These results are by the opinion of Roy *et al*. (2020) that artificial feed to replace live feed can reduce cannibalism and increase survival rates but affect on feed consumption in snakehead post-larvae. Furthermore, Chepkirui-Boit *et al*. (2011) stated that the growth performance, survival, and utilization of nutrients by fish larvae could be improved by giving artificial feed combined with natural food.

Conclusion

Combination of 50% dry silkworms and 50% artificial feed is the best combination of feeding in the rearing snakehead post-larvae.

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