

Growth Performance of Silkworms (*Tubifex* sp.) Cultivated Using a Dry Substrate

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Submitted: 31 May 2021; Revised: 27 September 2021; Accepted: 15 November 2021

ABSTRACT Silkworms are one type of live food that has intensified their cultivation activities in Indonesia. This study aimed to determine the effect of feeding/dried substrate in different doses on the growth performance of silkworms. The second aim was to determine the appropriate dose of dry substrate for silkworm cultivation. The study used a completely randomized design (CRD) with 4 treatments of dry media at a dose of 114 g/m² (D10), 228 g/m² (D20), and 324 g/m² (D30) dan without media as a control (D0). With 3 replications for each treatment. A silkworm starter was obtained from cultivation for 30 days. The research was conducted at Warung Benih Farm in Depok for 33 days. Data were analyzed using Analysis of Variance (ANOVA) and Duncan's further test with a confidence level of 0.05. The results showed that absolute weight growth, population growth, and the best productivity were obtained in the treatment of substrate addition with a dose of 324 g/m² (D30) with an absolute biomass growth of 48.67 g, population growth of 59458 individuals, & the productivity of 0.556 g/m² was/cycle. Most of the water quality observations are in the relatively optimal range.

Keywords: Absolute biomass; dry substrate; silkworm; population; productivity

INTRODUCTION

The cultivation of silkworms (*Tubifex* sp.) is one of the cultivation activities that have begun to be developed in the community. This is done to offset the high demand for silkworms and maintain the quality of harvested silkworms (Setyaningrum *et al.*, 2016). Nowadays, silkworms production can only meet 35-37% of market demand (Affi & Setia, 2017). The rest is still very dependent on a wild catch. Wild-caught silkworms have shortcomings, such as low quality and discontinuous availability (Yanti *et al.*, 2020). Silkworms harvested from the wild have a high possibility of carrying heavy metals and potentially zoonosis (Nurfitriani *et al.*, 2014). This happens because, generally, silkworms are found in ditches or rivers, which are usually polluted and contain heavy metals (Bintaryanto & Taufikurohmah, 2013; Singh *et al.*, 2010). From the availability point of view, wild-caught silk worms depend on the season. When many hatcheries are started in the rainy season, silkworms are challenging to obtain because they are carried away by water currents. This will also impact the selling price of silkworms (Yanti *et al.*, 2020). In the summer, the primary habitat of the silkworm, such as ditches and rivers, become dry, so silkworms are not found.

The technology for cultivating silkworms has been carried out in various ways. Starting from selecting the type of substrate to be used and the method of cultivation that will be carried out. The selection of the exemplary substrate can be an indicator of the success of silkworm cultivation because the quality and growth of silkworms are strongly influenced by the cultivation substrate used (Wenda *et al.*, 2018). There are also various cultivation methods, such as traditional paddy fields, apartment systems, and multi-storey shelving systems (Lestari *et al.*, 2021). Each of these technologies has its own set of benefits and drawbacks.

Cultivators usually plan their efforts around the availability of materials to use as a living medium for silkworms and the technology available in their local areas.

In this modern era, people, especially in the world of aquaculture, need something instantaneous to do business. Especially in the cultivation of silkworms, selecting the proper substrate is often a problem (Yanti *et al.*, 2020). In previous studies, substrate and dry feed have been obtained that can be packaged and stored for an extended period for cultivating silkworms. The feed is a formulation of several previous studies that have been carried out by Umidayati *et al.* (2020). The composition contained 40% tofu dregs, 25% fish silage, 25% fine bran and 10% mustard greens. These materials were dried at a temperature of 85 °C for 2 hours.

Furthermore, for the application of dry media, it must be precise in terms of the amount to be given. If there is a lack of silkworms, they will lack nutrients, inhibiting their growth (Mizwar *et al.*, 2021). Therefore, it is necessary to research the correct dry media dose to obtain optimal silkworm growth.

MATERIALS AND METHODS

Materials

The media used in this study was dry media from previous studies. The substrate composition is 40% tofu dregs, 25% fish silage, 25% fine bran and 10% mustard greens. These materials were dried at 85 °C for 2 hours. The nutritional content of the dry substrate used was 17.89% protein, 2.07% water, 15.94% ash, 8.71% fat, and 55.01% carbohydrates. The silkworm resulted from cultivation using a multilevel rack using fermented tofu dregs as a substrate and feed. The age of silkworms used was 30 days of culture. The equipment used as a cultivation container was a clear rectangular tray with a length of 35 cm, a width of 25 cm

and a height of 10 cm. On the left/right side of the tray, holes were made at the height of 5 cm from the bottom of the tray, and the hole diameter on the tray was 6 mm. The trays were arranged on a wooden rack with a length of 400 cm, a width of 35 cm and a height of 120 cm. pH was observed by an ATC pH meter with an accuracy of 0.01. Temperature and Dissolved Oxygen were measured using a mercury thermometer with an accuracy of 1°C and a DO meter brand Lutron-5510 with an accuracy of 0.01 mg/l.

Experimental design

The design of experimental was a completely randomized design (CRD) which included 4 treatments with 3 replications, as follows:

1. Without substrate/feed as control (D0).
2. Additional substrate/feed dose 114 g/m² (D10)
3. Additional substrate/feed dose 228 g/m² (D20)
4. Additional substrate/feed dose 342 g/m² (D30)

Research preparation

There were 12 plastic containers arranged on the rack, equipped with a storage tank at the lower part. The water in the storage tank container was circulated using a pump continuously during cultivation. The container that has been filled with water is put in the substrate according to the treatment. Silkworms are stocked in containers that are already filled with water and media. Before being stocked in the research container, the silkworms were quarantined for 7 days in the quarantine container to reduce bacteria and dirt on the silkworms. Furthermore, silkworms are stocked on trays with 100 g/tray stocking density.

Cultivation

Treatment during cultivation included additional substrate/feed according to treatment every 2 days. Everyday cleaning was carried out on the circulation holes and the water pump to unleash the flow of water and add water to the reservoir, which was reduced due to evaporation.

Harvesting

Harvesting was carried out at the age of 33 days of cultivation. The substrate and worms in the container were sieved. After that, the worms were put into a container. The surface of the worms was added 2 layers of gauze and allowed to stand for 120 minutes, and closed on the top to separate the worms and the substrate. It must be done until the worms are cleaned before weighing (Suryadin *et al.*, 2017).

Observation parameters

Absolute biomass growth

The total biomass of silkworms was calculated using the formula by Fajri *et al.* (2013) as follows:

$$W = W_t - W_o$$

Description : W = Absolute growth
W_t = Biomassa at stocking time
W_o = Biomassa at harvesting

Population growth

In addition, the population growth was calculated by taking a random sample in each treatment (Putri *et al.*, 2014). Calculating the silkworm population was done by counting directly from the sample of about 1 g. After that, it was converted to biomass weight in each tray (Hadiroseyani *et al.*, 2008).

Productivity

The productivity was calculated based on absolute weight. Productivity was calculated based on the formula of Lutfiyannah & Djunaidah (2020):

$$\text{Productivity} = \frac{(W_t - W_o)}{L}$$

Description : W_t = Final biomass (kg)
W_o = Initial biomass (kg)
L = Container area (m²)

Water quality

Observed water quality parameters included pH, temperature, and DO were carried out once every 7 days, while ammonia and nitrite were measured 3 times at the study's beginning, middle and end. A pH meter was measured using a pH meter, and DO was measured using a DO meter. The temperature was measured using a thermometer, while ammonia and nitrite were measured using spectrophotometry at the Test Laboratory of the Depok Ornamental Fish Cultivation Research.

Statistical analysis

The collected data were analyzed using Microsoft Excel 2013 and SPSS version 21. Several parameters were tested (i.e. absolute weight growth, population growth, and productivity) statistically analyzed by analysis of variance (ANOVA Single Factor) at the level of confidence of 95%. Furthermore, Duncan's test was carried out to determine the best treatment after knowing there were significant differences among treatments. Water quality was analyzed descriptively.

RESULTS AND DISCUSSION

Observations on absolute weight growth were conducted to determine the weight gain of silkworms that had been reared. Population growth was observed to determine the increase of individual silkworms and the dominant individuals in the rearing container. Productivity indicates an aquaculture business's success, calculated in units (kg/m²/cycle). Productivity is also related to the plan to develop

Table 1. Observation results of silkworm cultivation test parameters.

Parameters	Addition dose substrate/feed			
	D0(Control)	D10	D20	D30
Absolute biomass growth (g)	-29.67±14.22 ^a	17.00±5.29 ^b	34.33±7.09 ^c	48.67±6.03 ^c
Population growth (idv)	-8791±7973 ^a	34428±9721 ^b	36643±5380 ^b	59458±8741 ^c
Productivity (kg/m ² /cycle)	-0.339±0.163 ^a	0.194±0.060 ^b	0.392±0.081 ^c	0.556±0.069 ^c

Description: The mean values with different superscript letters on the same line show significantly different results (P<0,05).

a business area for silkworm cultivation. The results of observations in this study are shown in [Table 1](#).

Absolute biomass growth

The best absolute biomass growth was obtained at D30 with a growth of 48.67 g. Furthermore, in control, there was a decrease in absolute weight of 29.67 g. In the D10 treatment, the absolute growth was 17.00 g, followed by D20 with a weight of 34.33 g. The results of the analysis of the variance of giving different media doses have a significant effect on absolute weight growth (Sig=P<0.05). The growth of absolute weight can be graphically presented in [Figure 1](#).

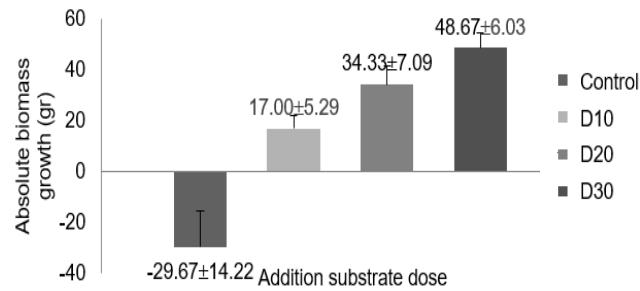


Figure 1. Absolute biomass growth of silkworm cultured for 33 days.

Population growth

The highest population growth of silkworms was found at D30, 59458 individuals, followed by D20 (36643 individuals), while the lowest population growth was found at D10, following a decline in the control population with a total of 8791 individuals. The analysis of variance showed that the dose of different media had a significant effect on the population growth (Sig=P<0.05).

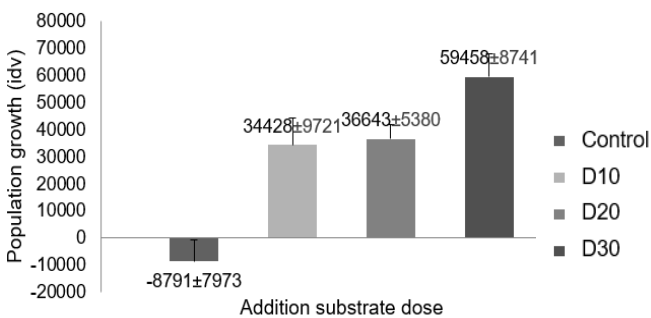


Figure 2. Population growth of silkworm cultured for 33 days.

Productivity

Productivity is calculated in units (kg/m²/cycle). In [Figure 3](#), the highest productivity is obtained at D30, 0.556 kg/m²/cycle. The lowest treatment was at D10 (0.194 kg/m²/cycle). The control in this study experienced a decrease in

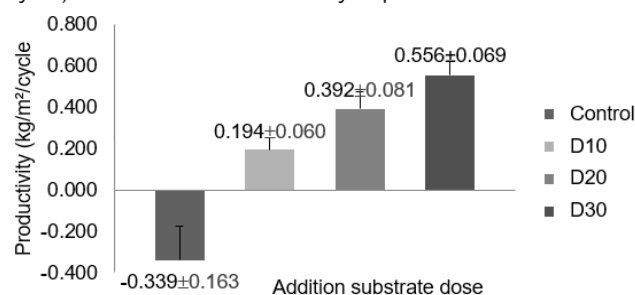


Figure 3. Productivity of silkworm cultured for 33 days.

productivity of as much as -0.339 kg/m²/cycle. The analysis of variance showed that giving different doses affected the productivity of the silkworms (Sig=P<0.05).

Water quality

In addition to observations on the growth of silkworms, water quality parameters were also an indicator to be observed. The results showed that qualities such as pH were in the range of 6.50-7.23, the temperature was in the range of 28-29 °C, dissolved oxygen was in the range of 2.4-4.6 mg/l, ammonia was in the range of 0.74-3.86 mg/l, while nitrite was in the low range of 0.01-0.04 mg/l. The observed water quality parameter values for 33 days can be seen in [Table 2](#).

Table 2. Observation value of water quality parameters during the study.

Parameters	Result of observation	Ideal range
pH	6.50-7.23	5.5-8.0*
Temperature (°C)	27-29	25-28**
Dissolved Oxygen (mg/l)	2.4-4.6	0.2-5.5***
Amonia (mg/l)	0.74-3.86	<3.6****
Nitrit (mg/l)	0.01-0.04	<1.5*****

Description: *. [Efendi \(2013\)](#) **. [Akhriil et al., 2019](#) ***. [Fadhulllah et al., 2017](#) ****. [Anggraini, 2017](#) *****. [Akinwole & Faturoti, 2007](#).

The water quality parameter values range between each treatment did not differ much. This is because each treatment uses the same water source and a closed recirculation system. The water stored in the reservoir is re-circulated in each cultivation container using a pump to distribute the water evenly. Absolute biomass growth

The highest weight growth was obtained in the addition of media with a dose of 30 g/container given every 2 days, with the absolute weight growth obtained being 48.67 g from the initial stocking of 100 g. This discovery contrasts sharply with the findings of a study by [Poluruy et al. \(2019\)](#), with an absolute weight growth obtained by 80% cultured for 45 days. The addition of different media doses gave a difference in the thickness of the substrate, which would be directly proportional to the growth of absolute weight. Silkworms need substrate and feed in their cultivation containers to breed. Furthermore, growth is due to protein and organic matter in the culture substrate, which is absorbed by silkworms ([Fajri et al., 2013](#); [Barades & Witoko, 2018](#)). The content of organic matter is obtained from bacteria decomposition by utilizing carbon (C) derived from molasses ([Syaputra et al., 2017](#)). The protein content in the media becomes a source of organic nitrogen utilized by microorganisms, and then the organisms become fed to the silkworms ([Akhriil et al., 2019](#); [Fachri et al., 2016](#)). The quality and source of protein largely determine the efficiency of protein absorption. In this study, the protein source used was a combination of animal and vegetable protein. According to [Hastuti et al. \(2016\)](#), combining several protein sources produces a better level of protein efficiency when compared to any single protein source.

Aside from providing a source of protein for silkworms to breed, the carbs and fats in feed can also provide energy

to sustain silkworm growth (Kusumorini *et al.*, 2017). The same thing was conveyed by Syahputra *et al.* (2020) that the carbon contained in the feed is helpful as an energy source, while nitrogen plays a role in supporting the growth of microorganisms. In control (D0), it was found that there was a decrease in absolute weight gain. This is because there is no additional substrate in the cultivation container. So there is an indication that the silkworm died in the container. Death can also be caused by the adult silkworm's growth peak, while the young silkworms cannot grow due to a lack of nutritional sources to breed (Kusumorini *et al.*, 2017). In addition, death can also be caused by insect larvae (*Chironomus*) which are competitors for silkworms. The presence of *Chironomus* will eat detritus and bacteria, and even silkworms that have just come out of their cocoons (Marian & Pandian, 1984). The inclusion of *Chironomus* in the cultivation container is due to the cultivation of silkworms in the open area, so it is easy for insect contamination to occur from the outside.

Population growth

The D30 treatment, which was the best treatment, had the highest individual growth results, with 59458 individuals/container. As a result, silkworms can thrive since the food elements in their environment are sufficient (Martudi *et al.*, 2017). Conversely, a lack of nutritional value will stymie the population's reproduction and growth (Syahendra *et al.*, 2016). The more the amount of feed given, the more the number of silkworms that breed. The food for silkworms to breed is from organic matter and decomposing bacteria.

The source of organic matter is obtained from the media given to the cultivation container. The growth of silkworms can also be seen on day 30, from observations it was found that many individual worms were small and the colonies were getting bigger in the rearing container. It can be seen that the silkworms have laid their eggs and hatched so that a silkworm colony appears, which is dominated by the small silkworms. Sampling results also show that at the beginning of cultivation, the number of individuals in 1 gr was 447 individuals/gr, while at harvest, the average value was 660 individuals/gr. From this, it can be seen that young silkworms begin to dominate, and adult silkworms, as starters, are already at their peak of growth. This discovery can be linked to the viewpoint expressed by Kusumorini *et al.* (2017) that the growth from the egg to the young worm that emerges from the cocoon is 10-12 days at a temperature of 24°C. Furthermore, at the age of 50-57 days, the silkworms can put their cocoons back.

At P0, there was a very significant individual decrease among other treatments. This is due to insufficient nutritional content supporting the growth process. The silkworms will consume the nutrients in the cultivation substrate first for survival and later for reproduction (Masrurrotun *et al.*, 2014). The absence of additional nutritional content causes the silkworm population to decrease. In addition, the reduced population can also be caused by the death of individuals already in the peak phase of their life cycle. It is explained by Putri *et al.* (2018) that there are 4 life phases in silkworms, namely the adaptation phase, the growth phase (log), the stationary phase, and the death phase Febrianti *et al.* (2020) added that the peak phase of silkworm growth is at 40 days, then it will decrease until 50 days. While topics that aren't

that dissimilar were also communicated by Johari (2012) that the life phase of silkworms from egg to adult is 37-50 days.

Productivity

Productivity is related to the yield of biomass obtained, so the pattern of the effect of productivity obtained is also relatively the same as the biomass obtained in each treatment (Sriwahyuni *et al.*, 2019). Based on the results of this study, the highest productivity was obtained at D30 with a value of 0.556 kg/m²/cycle. As the difference between cultivation yields using the traditional system, productivity is 0.5 litre/m²/month, while productivity is 1.2 litre/m²/month for the apartment system. Compared with research conducted by Umidayati *et al.* (2020), the productivity was 466 g/m²/cycle, slightly lower than the results of the research conducted. This is because the cultivation period in this study was 33 days, while in research conducted by Umidayati *et al.* (2020) just 21 days. Harvesting of silkworms should be done at the peak of average growth to obtain optimal results.

Water quality

The pH value in each cultivation container was still in normal conditions, which ranged from 6.50 to 7.23. Silkworms can live at a pH of 5.5-8.0. The pH value fluctuated with increasing cultivation time. However, fluctuations are still at a value that silkworms can still tolerate. Chilmawati *et al.* (2013) said that silkworms from the *Tubificidae* family have a water pH tolerance of 6-8 so that the pH conditions in the cultivation of silkworms are still able to support their survival.

The temperature range in the study was still in average conditions that could support the growth of silkworms, ranging from 27-29°C. The temperature fluctuations were also not too significant. Weather conditions cause fluctuations in temperature. This is because maintenance is carried out in an open area which causes temperature conditions in the maintenance container to be significantly affected by the temperature of the surrounding environment. Temperature is not a limiting factor for worms from the oligochaete family (Shafrudin *et al.*, 2005), but if the water temperature increases, it will affect the metabolic rate, and the need for oxygen will increase, as well as the toxicity of pollutants. The results of the observations showed that the temperature reached 29°C. The temperature is still in normal conditions, according to Pursetyo *et al.* (2011), because the temperature was between 28-30°C in his research.

Dissolved oxygen content is also one of the crucial parameters observed. Although silkworms grow in polluted areas with poor water quality in nature, in the cultivation process, water quality conditions are always maintained so that growth remains stable, and water circulation must be smooth so that oxygen needs are met (Ngatung *et al.*, 2017). The oxygen concentration in each container was relatively the same because the water source used was the same, and the circulation of water entering the cultivation container was always tuned to remain stable. Incoming water that circulates continuously raises oxygen levels and washes off hazardous contaminants in the media (Akhriil *et al.*, 2019). The lowest oxygen content is 2.4 mg/l, which is still above the threshold for silkworm cultivation's optimal

dissolved oxygen content. Silkworms may live in anaerobic circumstances for 48 days at a temperature of 2°C if oxygen levels are as low as 0.5 mg/l (Muliastari, 1993; Pardiansyah et al., 2014). However, to grow and develop, the oxygen level in the embryonic phase must be >2mg/l (Hildayanti, 2012).

The highest ammonia content reached 3.86 mg/l on day 19. This could be caused by the metabolism of silkworms and the addition of media to the rearing container and decomposition (Mizwar et al., 2021). This causes the ammonia content to increase. In addition, the high ammonia in the media is caused by the bacteria that carry out the nitrification process not yet playing an active role. The ammonia level recorded at the end of the study was reduced to 2.48 mg/l, which was attributed to the addition of water the day before the test sample was taken.

According to Anggraini (2017), the optimal value of ammonia is <3.6 mg/l, but in this study, it was found that the ammonia content reached 3.86 mg/l. This value is slightly higher than the quality standard for ammonia content. This shows that silkworms are not too disturbed by high ammonia levels in the media because they can still live in these conditions. The value of ammonia in water is always closely related to the pH value and also temperature (Boyd & Lichtkopler, 1979). So that the pH and temperature conditions in the rearing container are strived to be stable to avoid increasing ammonia levels. In addition, the high ammonia is thought to be caused by the increasing amount of substrate given to the rearing container. The given media contains various sources of protein in it. Protein compounds that do not decompose will increase the ammonia content (Anggraini, 2017). The nitrite content is still in the tolerable range of 0.01-0.04.

CONCLUSIONS

Based on this research, important conclusions can be drawn that will answer the research objectives, including that the difference in the dose of dried feed has a significant effect on the growth performance of silkworms ($P < 0.05$) with absolute biomass growth in the control that is -29.67 ± 14.22 , on D10 17.00 ± 5.29 , D20 34.33 ± 7.09 , D30 48.67 ± 6.03 . Population growth in the control -8791 ± 7973 , D10 34428 ± 9721 , D20 36643 ± 5380 , D30 59458 ± 8741 . Productivity in control -0.339 ± 0.163 , D10 0.194 ± 0.060 , D20 0.392 ± 0.081 , and D30 0.556 ± 0.069 . The best dry media dose is 30 g, which is given once every 2 days. With the growth performance, the absolute weight growth was 48.67 g, the population growth was 59458 individuals, and the resulting productivity was $0.556 \text{ kg/m}^2/\text{cycle}$.

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