Ginger (*Zingiber officinale* Roscoe) flour diet in gourami (*Osphronemus gourami*) hatchery with biofloc system

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Abstract. Ginger (*Zingiber officinale* Roscoe) is a supplement used to improve quality and productivity in fish farming. Various studies reported that ginger used in fish feed increased growth. This study aimed to determine if the addition of ginger flour increased the growth of gourami (*Osphronemus gourami*) fry in biofloc media. The study used gourami fish seeds placed in plastic tanks, each containing 20 fishes with three replications. The study was conducted in a biofloc system by adding ginger flour in feed at a dose of 5.63 g kg^{-1} feed (T1), 3.75 g kg^{-1} feed (T2), 1.88 g kg^{-1} feed (T3), and control (T0) for 90 days. Fish were fed 2 times a day with 3% kg^{-1} of biomass with a protein content of 33%. The results showed that ginger flour supplementation through feed could significantly increase growth in weight (WG) and length (LG), specific growth rate (SGR), feed conversion rate (FCR), and feed efficiency (FE) ($p<0.5$) in gourami seeds in a biofloc system. It was concluded that dietary ginger flour can be used to increase fish growth in aquaculture with a biofloc system.
Key Words: biofloc, ginger flour, growth, *Osphronemus gourami*.

Introduction. Ginger (*Zingiber officinale* Roscoe) has many benefits as a medicinal plant. Thus, ginger contains many bioactive compounds such as flavonoids, alkaloids, polyphenols, steroids, saponins, fiber, tannins, carbohydrates, carotenoids, vitamins, and minerals (Otunola et al 2010). Studies reported that herbs such as ginger have biological effects, promoting growth and being immunostimulant (Payung et al 2017). Gourami (*Osphronemus gourami* Lacepède, 1801) is one of the popular consumption fish in Indonesia. Market demand for gourami is quite high but its availability is limited.

To overcome this problem, it is necessary to intensify the cultivation of gourami. According to Manullang (2020), intensification is one alternative to increase fish production by increasing stocking density while utilizing limited land and having high productivity. However, intensification of cultivation through increasing stocking density can harm fish growth and can reduce water quality. Likewise, high density can lead to cannibalism and competition for space, oxygen, and feed which slows growth. In addition, intensive cultivation can cause the residual feed to accumulate and not decompose. To prevent adverse effects on intensive cultivation, maintenance with a biofloc system can be used.

Biofloc technology is a technique used in intensive aquaculture systems that can control pond water quality. The principle of biofloc is to balance carbon and nitrogen quantity in pond water. This system is gaining attention in aquaculture because of its potential to get high production from excess feed and metabolic waste. Biofloc works with the addition of carbon as an organic material for microbiology or aerobic bacteria to decompose and maintain the population of floc (Rijal et al 2020). In addition to obtaining additional feed to accelerate growth, additional supplements are needed so that growth becomes more optimal. Ginger flour *Z. officinale* is one of the supplements that can be used to increase fish growth (Manoppo, 2015). Ginger contains bioactive components such as oleoresin and gingerol essential oils which function to optimize the function of the digestive organs. Essential oils stimulate the secretion of digestive enzymes that promote growth. In addition, ginger increases appetite strengthens the stomach and improves digestion (Setyanto et al 2012)

Previous studies used herbs for fish farming, but only a few were applied in the biofloc system. Biofloc is a cultivation system that can maintain good water quality and, consequently, there is no need for water changes except to replace the volume of evaporated water, and it is comfortable for the fish that are kept (Salamah & Zulpikar, 2020). To support growth, a growth promoter of ginger flour is added through feed so that its growth is optimal. Although ginger flour is not directly important for biofloc, both biofloc and ginger flour supplements in feed have the same function to produce optimal growth of gourami fish. Therefore, this study aimed to determine the effect of feed supplementation with ginger flour *Z. officinale* to increase the growth of *Osphronemus gourami* in the biofloc system.

Materials and method. The research was conducted in March-June 2021 in the wet laboratory of the biofloc pond in Karangsari Village, Kembaran District, Banyumas Regency. The main study used *Zingiber officinale* ginger flour, gourami seed (2,68-2,93 cm), probiotic EM4, and molasses for biofloc.

The research used an experimental method with a Completely Randomized Design (CRD) consisting of 3 treatments, 1 control, and 3 replications. The study was conducted in a biofloc system. The treatment consisted in giving ginger flour in feed. It was considered that the optimum dose of ginger flour was 7 g kg^{-1} of feed (Manoppo, 2015). The treatment in this study was supplementation of ginger flour through feed in cultivation using a biofloc system. The control (T0) was only biofloc media without supplementation (T0, control). Ginger flour was used in this study with doses of 75%, 50%, and 25% of the optimum doses, respectively 5.63 g (T1), 3.75 g (T2), and 1.88 g (T3) kg^{-1} feed. The biofloc used relied on the results of research from Rijal et al (2020), who added 10 mL of EM4 probiotics and 40 mL of molasses for every 144L of water.

Research procedure. The study used 12 plastic tanks with a diameter of 40 cm, filled with 60 L of water. Each tank was filled with 15 gourami seeds, then acclimatized for 1 week, fed with normal feed (without treatment) 2 times a day at 07.00 in the morning and 17.00 in the afternoon WIB (Waktu Indonesian Bagian Barat) time. After acclimatization was completed, the seeds were measured in weight and length and these measured were considered as initial measures. The study was conducted for 90 days and water quality was maintained constants in terms of temperature and pH. During the 90-day study, water temperature and pH were constantly measured and if they were outside the normal limits, researchers brought them to the normal limits.

Ginger flour Z. officinale. Ginger was bought from the market and then cleaned under running water, cut into smaller pieces, air-dried, then blended using a blender and mixed into the feed. After the mixture was homogeneous, it was air-dried at room temperature. After drying, it was put it in a plastic bag and stored until use. This procedure followed Manoppo giodelines (2015).

Preparation of biofloc culture media. In each tank containing 60 L of water, 35 g of salt were added and then left for 24 hours. Then probiotic 3.93 mL and molasses15.7 mL were added, further left for 2 weeks until floc is formed and the color of the water changed to brownish green (Rijal et al 2020). After biofloc media was ready, the experiment begun.

Feeding. The feed used in this experiment was commercially feed produced by PT Mataram Sakti, Surabaya, East Java. The nutritional composition consisted of 33% protein, 5% fat, 4% crude fiber, 12% ash content, and 10% water content. The feed was given at 3% of the biomass weight and was given twice a day in the morning at 07.00 and afternoon at 17.00.

Growth measurement. The measurement of the growth of seed was carried out by measuring the weight and length of the fish at the beginning and end of the experiment. Weight was measured an analytical balance and length by a ruler. During the experiment, the amount of feed consumption and the number of seed deaths were calculated.

Measurements of weight (WG) and length gain (LG) followed Zonneveld et al (1991) guidelines by calculating the final weight (Wt) and length (Lt) reduced with the initial weight (Wo) and length (Lo): WG = Wt – Wo; LG = Lt – Lo. Feed efficiency was: FE = [(Wt-Wo): F] x 100%, feed conversion rate was FCR = F: [(Wt+D) -Wo], specific growth rate was: SGR = [(LnWt-LnWo): t] x 100%. WG and LG were the gain in weight (g) and length (cm) of fish after 90 days of rearing. Wt and Lt were the weight and length of the fish on weighing day t (after 90 days), while Wo and Lo were the initial weight and length of gourami fry (day 0). FE is feed efficiency, and it indicates if the feed given is efficient in increasing weight gain (%). F is the amount of feed (g) consumed. FCR is the feed conversion ratio, and it shows the amount of feed consumed to increase the weight of 1 kg of biomass. Water quality measurements such as temperature and pH were carried out at the beginning, middle, and end of the experiment.

Data analysis. Data analysis used the one-way ANOVA program in SPSS at the 95% confidence level (α = 0.05). If the F test shows a significant difference, then proceed with the Duncan Multiple Range Test (DMRT) test at a 95% confidence level to determine the statistically significant differences between treatments. Water quality data which includes water temperature and pH were analyzed descriptively.

Results and discussion

Growth of gourami. Talpur et al (2013) proved that adding ginger to died improved growth performance and feed efficiency of Asian sea bass, *Lates calcarifer* (Bloch). Supplementing ginger in fish diets may enhance growth and will signify a change in a healthy weight for an individual body in the bodyweight of fish seeds. Data regarding WG, LG, SGR, FCR, and FE are listened to in Table 1.

Table 1
Growth parameters of gourami (*Osphronemus gourami*) juveniles fed with ginger flour (% feed) for 90 days experiments

Parameter	*Treatment*			
	T0	*T1*	*T2*	*T3*
Weight gain (g)	7,19 ± 0,68^{a}	9,42 ± 0,39^{b}	9,02 ± 0,60^{b}	9,20 ± 0,20^{b}
Length gain (cm)	4,94 ± 0,20^{a}	6,48 ± 0,65^{b}	5,94 ± 0,64^{ab}	6,09 ± 0,58^{b}
SGR (%)	2,16 ± 0,09^{a}	2,44 ± 0,05^{b}	2,42 ± 0,07^{b}	2,43 ± 0,02^{b}
Total feed (g)	603,51	615,03	606,22	611,42
FCR	4,82 ± 0,42^{b}	3,87 ± 0,12^{a}	3,98 ± 0,23^{a}	3,94 ± 0,11^{a}
Protein level (%)	33	33	33	33
Feed Efficiency (%)	20,85 ±1,72^{a}	25,84±0,93^{b}	25,16 ± 1,46^{b}	25,41±0,72^{b}

Note: data are presented as mean ± SEM; different superscripts show significant differences (p<0.05).

The results of statistical analysis of variance (ANOVA) at 95% confidence level showed that the addition of ginger flour to the feed of gourami seed in biofloc system significantly increased weight and length gain ($p < 0.05$). The highest weight of *O. gourami* seed was obtained with a ginger flour dose of 5.63 g kg^{-1} of feed (T1), with an average of 9.43 ±

0.39 g, while the lowest mean was 7.19 ± 0.68 g in T0, control group. The highest increase in length was obtained by adding ginger flour at dose 7.5 g kg^{-1} of feed (T1) with an average was 6.48 ± 0.65 cm, while the lowest was 4.94 ± 0.20 cm (T0). This was assumed to be the effect of adding ginger flour to commercial feed on the growth of fish seeds by biofloc system.

Growth in weight and length. The results of this experiment showed that the addition of ginger flour with a dose of 5.63 g kg^{-1} of feed for 90 days was the optimal dose for weight and length gain of *O. gourami* seeds among the study treatments. Previous studies (Swain et al., 2018) reported that dietary ginger in juvenile striped catfish, *Pangasianodon hypothalamus* (Sauvage, 1878), for 90 days proved to significantly increase SGR, weight gain, and FCR ($p<0.05$). Furthermore, Talpur et al (2013) also reported a significantly higher growth rate after adding 10 g kg^{-1} of ginger in the feed to rainbow trouts. The increase in weight gain might be attributable to the presence of alkaloids, flavonoids, polyphenols, saponin, steroids, tannin, fiber, carbohydrates, vitamins, carotenoids, and minerals present in ginger, according to Otunola et al (2010) and Shirin et al (2010).

The herbal diets were reported to improve animal performance by stimulating the secretion of the digestive enzyme that could result in improvements in digestibility, stimulating the appetite, and increasing food consumption (El-Desouky et al 2012). Talpur et al (2013) reported that the ginger diet can function as an appetizer that can improve the performance of the digestive system including the digestibility of food which in turn produces energy to increase its growth. The recommended dose of ginger is between 5 and 10 g kg^{-1} feed for optimal growth, survival, and FCR of Asian seabass fish. At the time of the study, the treated seed appeared to be more active than the control.

Ginger not only contains protease and lipase enzymes but has several other components that can promote growth. Ginger flour contains protein and various vitamins. According to Mao et al (2019), the percentage of vitamin and protein content in ginger in dry weight is as follows: thiamine 0.035%, riboflavin 0.015%, niacin 0.045%, pyridoxine 0.056%, vitamin C 44.0%, vitamin A 30 IU, and vitamin C. B 10.02 mg. The total vitamins contained in ginger rhizome based on the percentage of dry weight is 44.15% and protein is 1.8 g. The content of vitamin C is also quite large (44%) so it can be a growth stimulant. As stated by Purwati et al (2016), vitamin C is not a source of energy but is needed by fish as a catalyst for metabolic processes in the body, for normal growth, survival, and reproduction. Gunawan et al (2014), stated that vitamin C is necessary for growth because vitamin C maintains iron atoms and maintains the hydroxylase enzyme in the biosynthesis of calories. Vitamin C can be absorbed quickly in the tissues where collagen is formed, namely in the skin, dorsal fin, cartilage, cartilage of the mouth, head, jaw, gill-supporting cartilage, and fish bones. Furthermore, Lamin et al (2018) proved that dietary ginger flour at a dose of 8 g kg^{-1} of feed can improve the profile and growth of koi fish. Dietary of *Z. officinale* can increase nutrient utilization, which is reflected in weight gain (WG), specific growth rate (SGR), protein efficiency ratio (PER), and feed efficiency ratio (FER).

Specific Growth Rate (SGR). The results of the 90-day experiment on the specific growth rate (SGR) of gourami seeds that were given the addition of ginger flour through their feed were analyzed using SPSS. The results of the statistical test of variance (ANOVA) with a 95% confidence level showed that the feeding mixed with ginger flour had a significant difference in SGR compared to the control. The highest specific growth rate (SGR) of *Osphronemus gourami* seed was obtained at a dose of 5.63 g kg^{-1} of feed (T1) with a mean of 2.44 ± 0.05 and the lowest was 2.16 ± 0.09, in the control group.

Hassanin et al (2014) reported that ginger diet had a significant ($p < 0.05$) increase in final BW, body gain, body gain %, and specific growth rate (SGR) % compared to control diets in the case of tilapia (*Oreochromis niloticus*). It could be concluded that supplementation of ginger in fish diets as an alternative to antibiotics and therapeutic agents, had a significant additive benefit in growth performance and immune status of fish compared to the control treatments.

Feed conversion rate (FCR). Feed conversion ratio (FCR) is the ratio between the amount of feed consumed and weight gain. The results of observations for 90 days can be analyzed by FCR on *O. gourami* fry during the addition of ginger flour through the feed.

The results of the statistical analysis of variance (ANOVA) with a 95% confidence level showed that feeding mixed with ginger flour through feed significantly increased the FCR of fish fry ($p < 0.05$). The lowest FCR was at the treatment dose of 5.63 g kg^{-1} of feed (T1) with a mean of 3,87 $\pm$ 0,12 and the highest was at the control (T0) with a mean of 84.43 ± 8.46. A good FCR is a low value, which means that a small amount of feed can increase the weight of the biomass.

Supplementation of ginger flour through feed in all treatments resulted in a lower FCR than the control, this showed that ginger flour affects FCR. The results of research conducted by Robiansyah et al (2018), reported that ginger flour given through feed to tilapia can produce significant FCR values in all treatments at a dose of 5.63 g kg^{-1} of feed. Furthermore, Jafarinejad et al (2020) stated that *Cyprinus carpio* carp fed a diet of 2 and 5% ginger which was cultivated for 56 days, increased FCR, growth, and final weight compared to the control diet. In addition to increasing FCR, a ginger diet can also improve immunity, health quality, and feed utilization efficiency. A low FCR value means that the amount of feed given is relatively low, and reduces water pollution during seed and fish rearing. The difference in the feed conversion value of each treatment showed differences in appetite.

Feed Efficiency. The results of the 90-day study on the feed efficiency of *O. gourami* seed with the addition of ginger flour through feed proved a significant effect. The results of the statistical analysis of variance (ANOVA) with a level of 95% confidence indicate that feeding mixed with ginger flour can significantly increase feed efficiency ($p<0.05$) on gourami seeds. The highest feed efficiency was achieved at a dose of 5.63 g kg^{-1} of feed (T1) with a mean of 1.53 ± 0.06 and the lowest was at control (T0) with a mean of 1.19 ± 0.11.

Previous studies showed that dietary ginger improved fish growth performance. Thus, Jafarinejad et al (2020) reported that ginger diet can improve the growth profile and efficiency of feed in tilapia. Supplementation of ginger can also increase the antioxidant capacity to respond to oxidative stress at the enzymatic and molecular level so that it can increase the body's immunity. This increase in immunity is considered to support an optimal growth profile in these experiments. Similar results were reported by (Alsaiad & Al-Zayat, 2019), who showed that the addition of ginger extract through feed on tilapia larvae for 16 weeks increased growth and feed efficiency value compared to control. Arief et al (2014) concluded that feed efficiency could be seen from various factors, one of which is feed conversion. A good level of feed efficiency will be achieved if it has a low feed conversion value.

Biofloc System. A biofloc system is a group of microorganisms that are united by bonds called biopolymers. Biofloc consists of algae, yeast, plankton, protozoa, and several other microscopic animals such as worms and others. Biofloc is formed by various organisms, and with the presence of bacteria in biofloc the degradation process of the organic matter becomes smooth, resulting in substances that are beneficial for plankton growth (Avnimelech, 1999). Biofloc can be defined as a cultivation system based on the principle of assimilation of inorganic nitrogen (ammonia, nitrite, and nitrate) by microbial communities (heterotrophic bacteria) in cultivation media which can then be utilized by cultured organisms as a food source (Widodo et al 2020).

The results of this experiment (Table 1) show that biofloc cultivation which was combined with supplementation of ginger flour through the feed can produce optimal growth performance at a dose of 5.63 g kg^{-1} feed (T1), indicating that biofloc cultivation with supplementation of ginger flour through the feed can increase optimal growth. The function of ginger flour as a growth promoter has been described previously, but the function of biofloc media has not been described. In the control (T0, biofloc only) and treatment with a lower dose of ginger flour (3.75 and 1.88 g kg^{-1} of feed or T2 & T3), the results were not optimum. With the addition of ginger flour to feed with this biofloc media, it is considered that biofloc media plays an important role in maintaining water quality.

This is following the report submitted by Ekasari (2009) who showed that the biofloc system maintained water quality because in this technology there were heterophilic bacteria that broke down organic and inorganic compounds in the water.

As stated by Schrader et al (2011), biofloc can suppress toxic compounds such as ammonia, and the growth of pathogenic bacteria so that the growth of cultured fish is better. Following the opinion of Hargreaves (2013), many advantages are obtained in shrimp farming with biofloc systems such as pH stabilization, reduction of ammonia content, and waste recycling (dead algae, feces, and food waste) because organic material can be used as feed for microorganisms and to form aggregates (flocs) that can be used as additional feed for tiger prawns (*Penaeus modonon*).

Support Parameter

Water quality. Good water quality can affect the maintenance of fish that are being cultivated. Water quality that is outside the optimum range will cause fish to experience stress so that they are more susceptible to disease. Therefore, in this study, water quality was maintained within the optimum range. The water quality observed in this study was water temperature and pH. Water quality is a factor that must be considered in fish maintenance. The results of the observation of water quality during the study can be seen in Table 2. It was observed that the ginger treatment affects the growth performance of gourami seeds, not water quality. This is in accordance with the research objectives.

Table 2

Water quality during 90 days of the experiment

Treatment	*pH*	*Temperature (C)*
T0	7.2-7.5	26-27
T1	7.6-7.9	26-27
T2	7.1-7.2	26-27
T3	7.3-7.6	26-27

Acidity levels (pH) and the temperature have a significant role in controlling the conditions of aquatic ecosystems. The pH and temperature measurements during the study was ranged from 7.1-7.9 and 26-27^{o}C. According to SNI (2000) in rearing gourami larvae, the optimal pH and temperature for rearing gourami fry are 6.5 – 8.5 and 25-30 ^{o}C. Consequently, both the pH and temperature in this study are within the normal limits of gourami cultivation.

Conclusion. Dietary of ginger in hatcheries during 90 days experiments in media biofloc increased growth of weight and length gain, specific growth rate, feed conversion rate, and feed efficiency of *O. gourami* seed. This shows that the ginger diet can be used in hatchery and the culture of gourami by biofloc system.

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Conflict of Interest. The authors declare that there is no conflict of interest.

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