Evaluation on feeding with sense leaves and enrichment on the conditioning of giant gourami (Osphronemus gouramy) broodstock

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Abstract. Spawning in the aquaculture of giant gourami is an important factor which determines the success of production because it supports the sustainability and improvement of the quality and quantity of giant gourami fry in aquaculture activities. The purpose of this study was to evaluate the feeding in the conditioning of giant gourami (Osphronemus gouramy) broodstock. The methodology carried out included the preparation of spawning ponds, broodstock selection, broodstock feeding, spawning, conditioning, nest inspection and egg collection, egg handling, egg hatching, and larval rearing. The eggs produced were 21,452 eggs with 18,714 fertilized eggs and 2,738 unfertilized eggs, and the Fertilization Rate (FR) value obtained during the study was 87.3%. Eggs that successfully hatched into larvae were as many as 16,238 while eggs that did not hatch were as many as 2,341 eggs, and the Hatching Rate (HR) value obtained was 87%. The water quality data obtained from the broodstock pond and the hatching pond included DO with a range of 5.88 – 6.11 mg/L, a temperature of 27.2°C – 28.2°C, and a pH ranging from 7.21 to 7.27.

1. Introduction

Aquaculture of giant gourami in ponds has long existed in Indonesia [1]. Giant gourami is a freshwater fish with high economic value due to the gourami aquaculture production that has not met a quite high market demand [2]. The growth of giant gourami tends to be slow compared to that of other freshwater fish. To reach the size of consumption with a minimum weight of 500g from 1g of fry, giant gourami requires maintenance time of more than one year. Despite this long maintenance time, giant gourami is favored by many people because of its savory taste, and also the meat is not mushy [3].

The total national production of giant gourami in 2015 was 113, 407 tons or fulfilled 70.75% of the 2015 national production target. National production of giant gourami in 2016 increased by 16.16% or into 149, 533 tons [4]. The production of giant gourami aquaculture will continue to increase along with high market demand and technological advances that most cultivators start to master. Spawning is an important factor and determinant of the success of production because it determines the sustainability and improvement of the quality and quantity of giant gourami in aquaculture activities. The hatchery technique of giant gourami (Osphronemus goramy) includes the preparation of spawning ponds, broodstock selection, spawning, egg harvesting, egg hatching and larval rearing, fish rearing, control of pests and diseases, harvest and post-harvest. The obstacles in the hatchery technique of giant gourami...
(Osphronemus goramy) consist of internal factors, namely fish biology and external factors, namely environmental factors, feed, water quality, and diseases [5,6].

Fish feed is one of the critical success components in fish farming. Based on the source, fish feed is divided into 3 groups, namely natural feed, supplementary feed and artificial feed. Natural feed is feed which is provided by nature and is already available in the pond, for example plankton (both phytoplankton and zooplankton), various aquatic plants (Azolla, mosses, algae, etc.) and aquatic animals [7][8]. Supplementary feed is feed which is added from outside the pond, for example leaves, household waste, and livestock waste. Artificial feed is feed which is prepared based on certain formulations aimed at obtaining feed with the required nutritional content and price [9,10].

Feeding with sente leaves (giant taro) is believed to increase survival, improve digestibility and suppress feed conversion. Sente leaves contain saponins, flavonoids, and polyphenols which can increase fish resistance [11]. However, the protein content of sente leaves is low, accounting for 4.96%; thus, feed pellets with high protein content are needed to meet the protein needs of giant gourami. Saponin compounds can stimulate immunity of giant gourami and can function as anti-fungal and anti-bacterial, but the administration of saponin compounds in excessive doses will be toxic to fish because saponins are poisons that can break blood grains [12]. Feeding with sente leaves will suppress the fat content so that it does not suppress the egg and sperm sacs. According to Fatin [13], flavonoid and polyphenolic compounds are able to inhibit fat oxidation. Based on the description above, an evaluation study was carried out on the provision of sente leaves as feed and on enrichment in the conditioning phase of giant gourami (Osphronemus gouramy) broodstock.

2. Materials and Methods
2.1. Broodstock Selection
Broodstock selection of giant gourami aimed to produce the breeding value of a population which can be increased through selection and to produce the best quality of fish in the hope that the selected fish can pass on their superior quality to their offspring [14]. The characteristics of broodstock ready to spawn can be seen in Table 1.

| Parameter      | Female          | Male       |
|----------------|-----------------|------------|
| Age            | ≥ 3 years       | ≥ 3 years  |
| Weight         | ≥ 2.5 kg        | ≥ 3 kg     |
| Forehead       | Not protruding  | Protruding |
| Bottom lip     | Thin            | Thick      |
| Pectoral Fin   | Black dark      | Bright white |
| Tail           | Pointed         | Flat       |

The male broodstock that had mature gonads were characterized by both ribs on the abdomen that were blunt. Meanwhile, the female broodstock were characterized by the bulging of the abdomen behind the pectoral fins and a change in the arrangement of the scales on the abdomen closer to the stretched pectoral fins [15]. Measurement of gonadal maturity level could also be done using a catheter to take eggs in the female broodstock’s body by inserting a catheter into the female giant gourami’s genital opening, and then the catheter was pulled so that the eggs entered the device. Afterwards, the catheter was removed from the genital hole. The eggs that had been obtained were then measured using millimeter paper. Eggs that had mature gonads had a size > 2 [16].

2.2. Broodstock Feeding
There were 2 types of feed for giant gourami broodstock, which were sente leaves and floating pellets (HI – PRO VIT). Feeding with sente leaves was 2% of broodstock biomass per day, while feeding with floating pellets was 1% of broodstock biomass per day in quantity. Feeding with sente leaves and floating pellets aimed to meet the protein needs of giant gourami [11]. Sente leaves as feed during the day was given to avoid itching caused by the oxalate content in sente leaf sap which can cause irritation.
in humans [17]. Taking sente leaves during the day can reduce the sap when cutting leaves due to sunlight exposure during the day on sente plants. In relation to this, heating has been the most common way to prevent itching caused by the calcium oxalate content in senteleaf sap. The feed pellets (HI – PROVIT) provided contained a number of nutrients which included protein of 31-33%, fat of 3-5%, fiber of 4-6%, ash content of 10-13%, and water content of 11-13%. Feeding with sente leaves would suppress the fat content so that it did not suppress the egg and sperm sacs. Feed pellets with high protein content could also stimulate gonadal formation and encourage spawning [18,19].

2.3. Spawning
There were 2 types of spawning systems used, namely the mass spawning system and the pair spawning system, using a male and female ratio of 1:3. This comparison ratio aimed to make spawning more effective because sperm cells were able to fertilize almost all ovum cells [20]. The mass system was a spawning system which was carried out en masse in one spawning pond with a total of 160 broodstock (40 males: 120 females). Meanwhile, the pair system was a spawning system which was carried out by making barriers made of nets and bamboo; 1 pond had ± 40 partitions with each partition filled with 4 broodstock (1 male: 3 females) BSN [21].

In general, there are several phases in spawning, namely copulation, ovulation, spermiation, and fertilization. Copulation is the process of adaptation of giant gourami brooders with a partner which is characterized by the process of fondling and chasing. Ovulation is the process of releasing eggs by the female broodstock in the nest that has been prepared by the male broodstock. Spermiation is the process of releasing sperm from the male broodstock. Fertilization is the process of fertilization of an egg by a sperm. Spawning is an activity of uniting male and female broodstocks who have mature gonads to fertilize eggs, in which the meeting of sperm cells with egg cells occurs [22]. Spawning was done in a concrete pond with a soil or mud bottom. The spawning of giant gourami was marked by a chase movement, and then the male and female broodstocks would simultaneously be in front of the mouth of the nest to spawn. Before the spawning process occurred, the male broodstock would first make an egg nest by arranging the fibers that had been provided. The spawning of giant gourami is not affected by season, or it occurs throughout the year [15].

2.4. Calculation of the Number of Eggs and Fertilization Rate
It was necessary to calculate the number of eggs resulting from the spawning process of giant gourami broodstock. As the eggs of giant gourami had a relatively large size, the calculation could be done manually, and the process was not difficult. In addition, fertilization rate is the percentage of fertilized eggs from the total number of eggs produced from the spawning process [15]. Observations were made based on the characteristics of fish eggs that were successfully fertilized and those that failed. Fish eggs that were successfully fertilized had a clear yellow color, while eggs that failed to be fertilized had a cloudy yellow color [16].

2.5. Hatching rate
Hatching rate is the percentage of the number of eggs that hatch from the total number of fertilized eggs [15]. Observations were made until no more hatched eggs were found. Hatched eggs were characterized by the movement of eggs that rotated on the surface of the water [23]. Meanwhile, eggs that did not hatch were marked with eggs that were cloudy yellow in color and sank to the bottom of the substrate. The calculation of the hatching rate was carried out by the formula [24].

2.6. Conditioning
Conditioning is a recovery stage for giant gourami broodstock. Conditioning was carried out after 3 months of spawning [25,26]. The giant gouramis in the spawning pond were separated first between males and females. After being separated, the giant gouramis were kept for 1 month to recover the broodstock. During conditioning, the feed pellets were added with some feed additives to support the recovery period of the giant gourami broodstock, or enrichment of the nutrients in the pellet feed given was conducted. The feed additives used can be seen in Table 2.
Table 2. Feed additives in feed conditioning

| Feed additives | Description |
|----------------|-------------|
| Egg yolk       | 30 eggs     |
| Vitamin E      | 60 capsules |
| Vitamin C      | 60 capsules |
| Corn oil       | 1.5 liter   |
| Water          | 1 liter     |

All these additives were mixed into 30 kg of pellets. The manufacture was conducted by mixing all the additives into the pellets, and after all the ingredients had been mixed, the pellets were dried until they were completely dry. Afterwards, the pellets could be put back into the feed container. Feeding on conditioning was different from the usual feeding. During conditioning, the feeding for the male broodstock was 3% of the fish biomass, specifically with 1% of sente leaves and 2% of pellets that had been enriched with nutrients. Meanwhile, for the female broodstock, the feeding was 5% of the fish biomass, specifically with 2% of sente leaves and 3% of pellets that had been enriched with nutrients.

Feeding with pellets was given more than feeding with the sente leaves as the protein content in pellets is greater than that in sente leaves. Protein is a substance needed for body maintenance, tissue formation, replacement of damaged body tissues, and the addition of body protein in the growth process [27]. Sente leaves were still given to meet the nutrition of giant gourami, and the excess protein in the fish's body would also be stored in the form of fat [28]. Vitamin E is one of the important micronutrients which affect the reproductive performance of fish. Vitamin E in feed can increase spawning success, egg fecundity and hatching rate, larval survival, gonadosomatic index, and vitellogenesis [29], while vitamin C in feed is effective in accelerating growth and maintaining fish survival [30]. Corn oil functions as an antioxidant which protects fat and prevents the oxidation process, so that the egg formation process can run normally [31]. Egg yolks are widely known to have a complete and good composition of essential amino acids so that when used in feed, they can stimulate fish growth [32].

3. Results and Discussion

3.1. Result

3.1.1. Fertilization Rate (FR)

Egg counting aimed to count the total number of eggs in one nest. Eggs were counted manually using a plastic spoon. At the time of counting eggs, egg sorting was also carried out which aimed to separate live and dead fish eggs. According to Harahap [33], eggs that die occur because the eggs are not fertilized. Live eggs would be bright yellow, while dead eggs would be milky white. The dead and live eggs were counted as a whole to determine the Fertilization Rate.

Table 3. Fertilization Rate (FR)

| Pond          | Eggs produced | Fertilized Eggs | Unfertilized eggs | FR (%) |
|---------------|---------------|-----------------|-------------------|--------|
| KI. 1. KM 7   | 3118          | 2555            | 563               | 82     |
| KI. 3 (en Masse) | 3285        | 2440            | 845               | 74     |
| KI. 1. KM 18  | 6820          | 5761            | 1059              | 84     |
| KI. 1 KM 9    | 4099          | 3942            | 157               | 96     |
| KI. 1 KM 1    | 4130          | 4016            | 114               | 97     |
| Total         | 21,452        | 18714           | 2738              | 433    |
| Average       | 4290.4        | 3742.8          | 547.6             | 86.6   |

Table 3 shows data on the fertilization rate in each pond with a fertilization rate of 86.6%, an average of eggs produced of 4290.4, an average of fertilized eggs of 3742.8, and an average of unfertilized eggs of 547.6.
3.1.2. Hatching Rate (HR)
The eggs of giant gourami experienced a mortality rate of 8 – 21% in the process of egg hatching and larval rearing. Egg mortality was caused by poor egg handling.

Table 4. Hatching Rate (HR)

| Pond                | Fertilized Eggs | Hatched Eggs | Unhatched Eggs | HR (%) |
|---------------------|-----------------|--------------|----------------|--------|
| KI. 1. KM 7         | 2555            | 2330         | 225            | 91     |
| KI. 3 (en Masse)    | 2440            | 2074         | 366            | 85     |
| KI. 1. KM 18        | 5761            | 4557         | 1204           | 79     |
| KI. 1 KM 9          | 3942            | 3626         | 316            | 92     |
| KI. 1 KM 1          | 4016            | 3696         | 320            | 92     |
| Total               | 18714           | 16283        | 2431           | 439    |
| Average             | 3742.8          | 3256.6       | 486.2          | 87.8   |

Table 4 shows data on the hatching rate in each pond with a hatching rate of 87.8%, an average of fertilized eggs of 3742.8 eggs, an average of hatched eggs of 3256.6, and an average of unhatched eggs of 486.2.

3.1.3. Water Quality
Table 5. Average data of water quality measurement results every week

| Parameter           | Broodstock Pond 1 | Broodstock Pond 3 | Fiber Hatching Tank |
|---------------------|--------------------|--------------------|---------------------|
| Temperature (°C)    | 27.3               | 27.2               | 28.2                |
| pH                  | 7.26               | 7.21               | 7.27                |
| DO (mg/L)           | 5.89               | 5.88               | 6.11                |

Table 5 shows the average data on water quality measurement results for the broodstock pond 1, broodstock pond 3, and fiber hatching tank including temperature, acidity (pH), and dissolved oxygen (DO) each week.

3.2. Discussion
Pond preparation for the giant gourami broodstock began with cleaning the pond to remove pests and dirt. The clean pond was then dried for 3-7 days, and afterwards the bottom soil of the pond was turned over to reduce the organic matter content at the bottom of the pond and to seal the pond embankment from leaking [34]. Liming the pond was done using quicklime at a dose of 50-150 gr/m². Liming was carried out with the aim of improving the acidity (pH) of the bottom soil of the pond and maintaining it in a stable condition; it also functioned as a disinfectant and provider of nutrients needed by phytoplankton [35,36].

The criteria for broodstock, ready for spawning, referred to SNI [21]. Giant gourami broodstock were also selected based on the level of gonad maturity. Visibly the male broodstock that had mature gonads were characterized by both ribs on the abdomen that were blunt [15]. Feeding giant gourami broodstock used 2 types of feeds, sente leaves and floating pellets (HI – PRO VIT). Feeding with sente leaves was 2% of broodstock biomass per day, while feeding with floating pellets was 1% of broodstock biomass per day in quantity. Feeding with sente leaves and floating pellets aimed to meet the protein needs of giant gourami [11].

The floating pellets given were 3-4 mm in size with 31-33% protein content, 4-6% fat, 3-5% fiber, and 9% water content. Feeding with sente leaves will suppress the fat content so that it does not suppress the egg and sperm sacs. Water is an important factor in aquaculture activities, so it must be in optimal conditions in terms of quality and quantity [37]. Water quality management in spawning activities is expected to provide optimal environmental requirements for the growth and reproduction of giant gourami. Sampling of water in the spawning ponds and larval hatching tanks was carried out in the
morning and evening. Some water quality parameters that were measured were temperature, dissolved oxygen (DO) and acidity (pH). The average data from the results of water quality measurements can be seen in Table 1.

The results of water quality measurements in broodstock ponds and fiber hatching and rearing tanks met the criteria for good water quality. According to SNI [21], the optimal temperature for spawning ponds ranged from 25 - 30°C, while in hatchery and rearing tanks it is 29-30°C. The optimal degree of acidity (pH) for spawning pond ranged from 6.5 to 8.0, while in hatchery and egg rearing tanks it was 6.7 to 8.6 [38,39]. According to Sulistyo [40], the optimum dissolved oxygen content was 4-7 mg/liter.

Fertilization rate is the percentage of fertilized eggs from the total eggs produced from the spawning process [15]. Fertilized eggs were separated from unfertilized eggs for further egg hatching and larval rearing. Fertilized eggs would be bright yellow in color, while unfertilized eggs would be milky white. The dead and live eggs were counted as a whole to determine the fertilization rate. The data of fertilized eggs can be seen in Table 2.

Giant gourami eggs which were successfully fertilized were as many as 18,714 eggs out of a total of 21,452 eggs with an average fertilization rate (FR) value obtained during the study of 87.3%. This indicates that the quality and number of sperm were good enough to fertilize eggs. According to Arfah [15], fertility is strongly influenced by the quality of sperm, eggs, media and human handling. Giant gourami eggs would then hatch after 36-48 hours. The eggs that hatched would become larvae that had an egg sac (yolk sac) and swam upside down with the belly facing the surface of the water. The success of hatching carp eggs was influenced by several internal and external factors. The internal factor referred to the quality of eggs produced from the spawning process. Meanwhile, the external factors referred to the treatment during the hatching process and the conditions of the hatchery environment which included the condition of the hatching container and the quality of the hatching water [41].

4. Conclusion
The results of the evaluation study of feeding on the conditioning of giant gourami broodstock (Osphronemus gouramy) started from the preparation of broodstock ponds, broodstock selection, spawning, feeding, conditioning, nest inspection and egg collection, egg handling, egg hatching, larval rearing, and larval handling. 21,452 eggs were produced from 5 nest samples with an average of 4,285 eggs per nest. Fertilization rate reached 87.3% with a hatching rate of 87%. The average water quality data in broodstock ponds included the temperature of 27°C, pH of 7.2, and DO of 5.8 mg/l, while in hatching fiber and larval rearing tanks, the temperature was 28°C, pH was 7.27, and DO was 6.11 mg/l.

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