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The Effect Of Concentration And Length Of Soaking Solution Of Brahman Cow Testic Flour (Bos Indicus) On The Formation Of Male Sex In Sangkuriang Catfish Larvaes (Clarias Gariepinus)

E. Yusni[1]*, Aprik[2]

[1*,2]*Departement Aquaculture Reesource, Faculty Agriculture Universitas Sumatera Utara, Medan Indonesia*

***Corresponding Author:** E. Yusni*
**Departement Aquaculture Reesource, Faculty Agriculture Universitas Sumatera Utara, Medan Indonesia*

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Abstract

The aim of this research was to detect the effect of giving Brahman cattle testicle flour on the achievement of the male catfish species produced. This research was conducted at the Fish Seed Center (BBI) and the Fish Pest and Disease Laboratory in the city of Medan. The method used in this research was a randomized block design (RAK) method by collecting testicular samples of Brahman cattle at the Tanjung Leprosy slaughterhouse in Medan and fish samples in Fish Seed Center (BBI). This research was carried out from February to August 2023, with samples of 270 catfish larvae (*Clarias gariepinus*), and 500g of Brahman cattle testicles (*Bos taurus indicus*). It is made into flour as a soaking treatment with a concentration of 50ppm, 60ppm and 70ppm and soaking is carried out for 12 hours, 18 hours and 24 hours. The results of this study stated that the highest weight in the 70 ppm concentration treatment (P3TS3) with 24 hour soaking resulted in a catfish weight of 1.856±0.094 gr and the lowest 1.673±0.031 gr was found in the 50 ppm concentration treatment with 12 hour soaking (P1TS1), while the highest length was in the 70 ppm concentration treatment with 24 hour soaking (P3TS3) it was 1.851±0.053 and the lowest in the 60 ppm concentration treatment with 18 hour soaking (P1TS2) was 1.946±0.063. The highest feed conversion ratio was found in the 70 ppm concentration treatment with 24 hour soaking (P3TS3) of 1.946±0.088 and the lowest was in the 70 ppm concentration treatment with 18 hour soaking (P3TS2) with a result of 1.830±0.174. For fish survival, the highest was in the treatment with a concentration of 60 ppm at 18 hours of soaking (P2TS2) and in the treatment with a concentration of 60 ppm at 24 hours of soaking (P2TS3) with a yield of 93% for both, while the lowest was in the treatment with a concentration of 60 ppm with 24 hours of soaking. (P2TS3) with a total of 70%. The results of the number of masculinated fish (male sex formation) were found in the treatment with a concentration of 50 ppm with 24 hour soaking (P1TS3) and the treatment with a concentration of 60 ppm with 12 hour soaking (P2TS1) produced 79% males, followed by the treatment with a concentration of 60 ppm with 18 hours of soaking. hours, (P1TS2), 60 ppm concentration treatment (P2TS3) with 24 hour soaking and 60 ppm concentration treatment (P2TS2) with 18 hour soaking. Meanwhile, the highest total of female catfish was found in the treatment with a concentration of 50 ppm and 24 hour soaking

producing 21% females and the lowest was at a concentration of 60 ppm (P2TS3) soaking for 24 hours.

Key: Testis flour, catfish, male sex.

Introduction

Brahman cattle were developed in the United States, Gulf region, between 1854 and 1926. American Brahmans include Zebu breeds of Kankrey, Ongole, Gir, Krishna, Hariana, and Bhagari. This breed of cattle, which originally developed in the United States, has now spread widely in both tropical and subtropical areas, namely in Australia and also in Indonesia (Sugeng,1998). The characteristic of Brahman cattle is that they have a large hump and loose skin, wattle under the neck and a wide belly. with many folds (Sakir, 2017). Long ears Brahman cattle were developed in the United States, Gulf region, between 1854 and 1926. American Brahmans include Zebu breeds of Kankrey, Ongole, Gir, Krishna, Hariana, and Bhagari. This breed of cattle, which originally developed in the United States, has now spread widely in both tropical and subtropical areas, namely in Australia and also in Indonesia (Sugeng, 1998). The characteristic of Brahman cattle is that they have a large hump and loose skin, wattle under the neck and a wide belly. with many folds (Sakir, 2017). Long ears hanging and pointed.
This cow is the best type of beef cattle to develop, its carcass percentage is 45%. There are two cow testicles, under normal circumstances both testicles are the same size, located in the prepubic area, encased in the scrotal sac and suspended by the funiculus spermaticus which contains elements carried by the testes in their movement from the abdominal cavity through the inguinal canal into the scrotum (Toelihere, 1981). Cow testicle flour is a natural ingredient that is often used in the fish breeding process. Based on several research results using cow testes, it shows that cow testes contain very high levels of the natural testosterone hormone. Apart from that, cow testicle flour is easy to obtain, the price is relatively cheap and the size is large (Hidayani, 2016). Cow testicles contain the natural testosterone hormone which can be used in the masculinization process, namely the effect of changing from female to male.

Cow testicles are also easy to obtain, relatively cheap, and large in size. To use it, it needs to be made into flour, so it doesn't rot quickly (E.Yusni et al 2021), doesn't reduce water quality and is easy to store. Cow testicle flour is a natural ingredient that is often used in the fish breeding process. Based on several research results using cow testes, it shows that cow testes contain very high levels of the natural testosterone hormone. Apart from that, cow testicle flour is easy to obtain, the price is relatively cheap and the size is large (Muslim 2011). Administration of hormones derived from cow testicles in the early phase of gonad growth when sexual differentiation has not yet been directed. However, if certain ingredients are intervened with, such as cow testicle flour, gonad development can proceed in the opposite direction as it should (Zairin Jr 2002). Cow testes contain the natural testosterone hormone which can be used in the masculinization process, namely the effect of changing from female to male (Mantau. , 2013.) . Another advantage of beef testicle flour is that it contains organic ingredients (nutrients) which are useful for the fish's body, does not cause physiological stress on the fish so it does not cause death to the fish, is environmentally friendly and safe for consumers (Rizal Akbar Hutagalung, 2020). Apart from that, testicles are a local resource that is easier to obtain (Muslim, 2010). Because these cows live in tropical temperatures, experts have tried to conduct research using cow testicles to produce male fish from various species, one of which is about the effect of male cow testicles on male sex in Tilapia fish with a male population of around 80% compared to female fish. (E. Yusni et al., 2020). Sangkuriang catfish have fast growth, are relatively resistant to disease, and the technology for cultivating Sangkuriang catfish is relatively easy for the community to master. Male fish are an important factor in cultivating Sangkuriang catfish, because in their development male seeds have great advantages in spurring fish production. faster, shorter harvest period, and efficient in utilizing feed as a source of growth energy (Rosmaidar et al. 2016).

To obtain superior male fish seeds, studization can be carried out or also known as sex reversal, as a technology that reverses the direction of sexual development to the opposite. Cow testes are a waste that is difficult to grind because it is rarely used in the slaughterhouse industry. Cow testicles function as producers of spermatozoa, male sex cells and can secrete hormones (testosterone). Cow testicle flour is a natural ingredient that is often used in fish maleness process (Setiawan et al., 2017). Based on several research results using cow testes, it shows that cow testes contain very high levels of the natural testosterone hormone. The process of absorbing hormones into the body of Sangkuriang catfish seeds occurs through a diffusion process, from the

insan then through the blood vessels the hormones will flow to the organs and nervous tissue. Arriving at the target cell, the incoming hormone binds to receptors located in the spermatozoa sac (Murni, 2015). Estrogen hormones, such as estrone, estriol, estradiol, and ethinylestradiol can be used or have a directing effect on sexual differentiation into females (feminization) (feminization). Donaldson and Benfey 1987., Ganong, 1995) and androgen hormones, such as androstenedione, ethinyltestosterone, methyltestosterone, and testosterone propionate which can be used or have the effect of directing sexual differentiation into males (masculinization) (Hasbi and Gustina, 2018., Aritonang, 2020) . The cow's testicular hormone testosterone can have an effect on gender, which is called reversal, which is a way of reversing the direction of sexual development in fish that should be female, leading to the development of their gonads to become male or vice versa (Zairin, 2002, Rosmaidar et al., 2016). The process of sexual differentiation in teleosts is gradual and unstable. Therefore, masculinization should be carried out at the age of 6-10 days after the eggs hatch and a maximum of 17-19 days (Suyanto, 1994).

This technique is carried out when the gonads (genitals) of the fish have not clearly differentiated between male and female at the time of birth. hatch (Ridwan, 2022). Cow testicles are easy to obtain, relatively cheap, and large in size, but they need to be processed into flour, so they do not rot quickly, do not reduce water quality, contain good nutrition and are easy to store and easy to obtain (Muslim, 2010). The reaction of the testosterone hormone from cow testes will influence the male gender of fish (Ariyanto et al., 2016). The younger the fish, the greater the chance of forming male sex (Shapiro., 1987). This is similar to tilapia where the growth of male red tilapia is faster than female tilapia (Jangkaru and Asih, 1988), generally can be produced commercially using sex reversal techniques using the hormone methyltestosterone (Adel et al., 2006). However, along with its development, it is feared that the use of synthetic hormones will have a negative impact on food safety and environmental sustainability (Bartet et al. 2003). For this reason, an alternative step in finding a replacement for synthetic hormones is to use natural compounds. Natural compounds have the advantage of being easily decomposed in the body, causing few side effects, and reducing operational costs (Hidayani et al., 2016). It is hoped that the use of compounds from natural ingredients can be easily applied at the fish farmer level to make it more effective and efficient (Wiryowidagdo 2005).

Fish sexual differentiation varies greatly depending on the species. The application of genital direction techniques is generally carried out on catfish when the larvae are 3-5 days after hatching, the most sensitive time for masculinization (differentiation period). The success of changing sex is not only determined by the type and dose of hormone used, but is also influenced by the length of time the hormone is given, therefore information is needed regarding the optimum length of time for soaking the seeds in a solution of cow testicle flour to change the female sex of catfish. Apart from that, male fish need to maintain their growth and this cow testicle flour is the answer to increasing male genitalia. Therefore, to prove this, this research needs to be carried out.

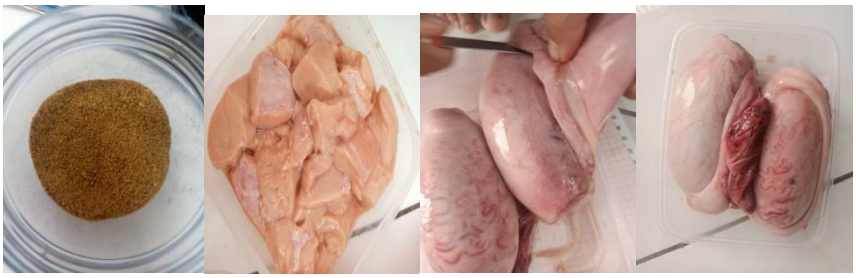
Materials and Methodology

Time and Place of Research

This research was carried out from March to May 2023. Fresh cow testes were obtained from the Tanjung Kusta Slaughterhouse (RPH), Medan City. The preparation of cow testicle flour solution and fish rearing were carried out at the Pest and Disease Laboratory of the UPT Fish Seed Center (BBI) of the Medan City Food, Agriculture and Fisheries Administration.

Preparation for Making Cow Testicle Flour

The cow testes used in this research weighed 250-500 g. The cow testes came from Brahman cattle (Bos indicus) obtained from the Tanjung Leprosy Slaughterhouse (RPH). Fresh cow testicles obtained from slaughtering at the slaughterhouse are skinned, split and cut into pieces, then chopped until smooth, the pieces of cow testicles are put into the oven at a temperature of 60 oC for 24 hours, the dry testicles are removed from the oven and ready to be used. grind it then put it in a porcelain bowl until it looks like flour, then sift it using a fine sieve. Place the finely ground testicular flour in an airtight jar and close tightly and ready to use, before use, store in the freezer (Setyawan, 2017).



a) b) c) d)

Figure 1. a) Cow testicles still intact, b) Cow testicles skinned, c) Cow testicles chopped, d) Cow testicle flour

Preparation of Materials and Fish Samples

A total of 27 jars with a diameter of 19 cm and a height of 18 cm were used as a place for soaking larvae, while 27 units of buckets with a diameter of 55 cm and a height of 42 cm were used for rearing fish. The samples of catfish (Clarias gariepinus) larvae used were 6 days old, as many as 270, each bucket contained 10 fish with 3 replications, then Brahman cattle (Bos indicus) testicle flour was ready to be used, for growing catfish (Clarias gariepinus), given food commercial. To observe the sex of males, methylene blue, acetocarmine solution, 70% alcohol solution and a microscope are used.

Preparation of Test Solutions and Soaking

A total of 50 ppm of finely ground beef testicle flour was put into a test tube then diluted with 2.5 ml of 70% alcohol. Each treatment was carried out with the same dilution, left for 1 hour, after that 10 fish were put into each jar by soaking. 50ppm for 12 hours with the symbol (P1TS1) after that the fish were moved to the rearing area, and the same was done for all treatments. For 18 hour soaking, the symbol (P1TS2) is given for 24 hour soaking (P1TS3), with treatment 1. Meanwhile, for the treatment, 60 ppm soaking for 12 hours is given the symbol (P2TS1), 18 hour soaking (P2TS2), and 24 hour soaking (P2TS3), . This treatment was the same as for treatment 2. And for the 70 ppm soaking treatment for 12 hours, the symbols were given as (P3TS1), 18 hour soaking (P3TS2) and 24 hour soaking (P3TS3) for Treatment 3. All cow testicle flour was carried out with the same dilution of 2, 5ml alcohol (70%) following the method (Rosmaidaret al., 2016). Each treatment was carried out with 3 replications for a total of 270 fish.

Feeding Fish

For 6 day old larvae, they were fed artemia for 10 days because the fish had not yet completed their growth. After ten days the fish were fed tubifex until the 30th day, this was done because the fish's mouth openings had begun to appear. For days 30 to 60 the fish are fed pellets, with a feed size of 0.2mm. Feeding was 3% of the fish's body weight, this treatment was given until the end of the study.

Fish Gender Check.

Examination of the sex of the fish was carried out when the fish were 60 days old or during the duration of this research. The method used in this research follows the method of Zairin (2002), namely starting from dissecting the fish's stomach using a scalpel, then the fish gonads are taken using tweezers and placed on a glass object, then finely chopped and added ± 2 to 6 drops of acetocarmine solution. then covered using a cover glass and observed under a microscope at 100 times magnification.

Percentage calculation

The sex of male catfish (Clarias gariepinus) follows Zairin (2002) that the calculation formula is as follows:

% Males = Number of Males/ Total number of fish x 100%
% females = Number of Females/Total number of fish x 100%

Fish Growth

Fish growth can be obtained by calculating the increase in fish weight from each treatment weighed during the research, according to Aritonang (2020) using the following formula
Weight growth: W = Wt –Wo

Information:
W: Fish growth (grams)
Wt: Average weight of individuals at the end of rearing (grams)
Wo: Average weight of individuals
Length growth: L = Lt – Lo

Information:
L: Length growth (cm)
Lt: Average individual length on day t (cm)
Lo: Average individual length on day 0 (cm)
Feeding conversion ratio (FCR) According to Effendi (1997), the feed conversion ratio can be calculated using the formula:
FCR = F / Wt−Wo

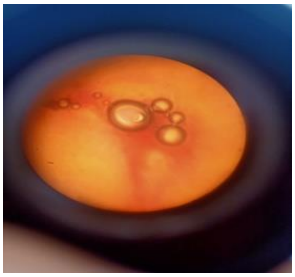
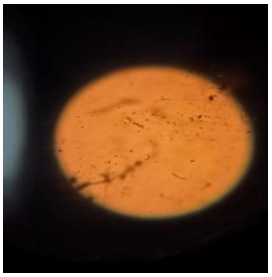
Information:
To calculate the feed conversion ratio from fish aged 30 days to 60 days during the research.
FCR: Feed conversion ratio
F: Amount of feed given (g)
Wt: Average weight at the end of the study (g)
Wo: average weight at the start of the study (g)

Survival Rate follows Effendi (1997) as follows:
SR (%) = Nt / No x 100%

Information:
SR: Fish survival during the experiment
Nt: Number of fish at the end of the experiment (tails)
No: Number of fish at the start of the experiment (tail

Data Analysis
Data was generated by ANOVA using statistical social science (SPSS) with a randomized block design, and data was taken from weight, length of fish, feed conversion ratio and fish survival during the study.

Results
The results of the research were carried out for 60 days, where soaking was done after 12 hours, then the fish were transferred to the rearing area, as well as soaking for 18 hours and 24 hours, 30 days for soaking and 30 days for growth. After 60 days, all fish were checked for sex as shown in Figure 2.


a) Fish genitals b) Male fish genitals c) Female fish genitals
Figure 2: Examination of fish genitalia

Data collected for fish rearing, both weight and length, were taken after the fish were fed pellets, namely on days 30 to 60, as well as feed conversion ratio data (Table 1). The effect of soaking cow testes shows a real difference on the weight and length of the fish and also on the feed conversion ratio as in Table 1. The average weight of fish treated with 50 ppm cow testicle flour and 12 hours soaking (P1TS1) is 1.688 ± 0.021 gr smaller than with 18 hour soaking (P1TS2) was 1.757 ± 0.020 gr and 24 hour soaking (P1TS3) was 1.756 ± 0.088 gr, and in the 60 ppm treatment with 12 hour and 18 hour soaking ((P2TS1) and (P2TS2) there was no difference in fish weight, namely, 1.877 ± 0.014gr and 1.844±0.041gr, the highest in 24 hour soaking (P2TS3) namely 1.904 ±0.015gr, and in the 70ppm treatment with 12 hour soaking (P3TS1) it was 1.735±0.012gr smaller than

18 and 24 hour soaking (P3TS2) and (P3TS3), namely 1.914 ± 0.024 gr and 1.988 ± 0.013 gr, can be seen in Figure 3 below.

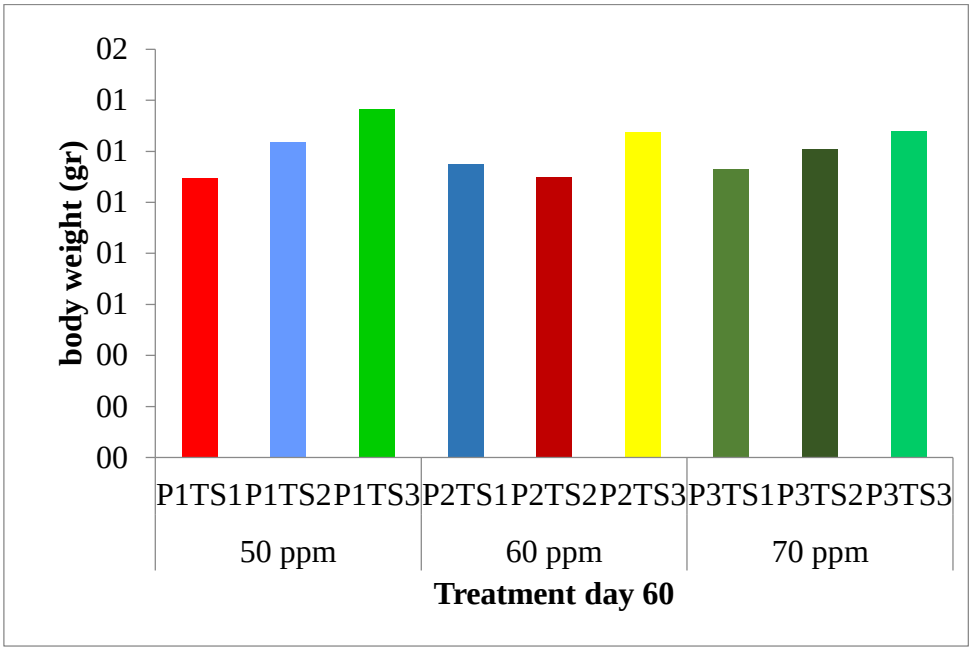


Figure 3 Differences in fish weight during the 60 days of research

Table 1: Mean and standard error of body weight, body length and feed conversion ratio on day 30 to day 60 with 3 different treatments

Treatment	Body weight (gr) (Day)			Body lebgth (cm) (Day)			FCR (Day)		
	30	40	60	30	40	60	30	40	60
P1TS1	1.673^{a} ±0.031	1.179^{a} ±0.030	1.087^{ab} ±0.055	1.906^{a} ±0.055	1.100^{a} ±0.038	0.891^{a} ±0.028	0.575^{ab} ±0.008	1.195^{cd} ±0.064	2.114^{ab} ±0.008
P1TS2	1.856^{a} 0.094	1.180^{a} ±0.091	1.153^{ab} ±0.066	1.946^{a} 0.063	1.180^{a} ±0.055	0.956^{ab} ±0.048	0.536^{a} ±0.013	1.276^{d} ±0.109	2.048^{ab} ±0.109
P1TS3	1.669^{a} ±0.072	1.221^{ab} ±0.079	1.335^{b} ±0.090	1.873^{a} ±0.031	1.041^{a} ±0.034	1.040^{ab} ±0.051	0.575^{ab} ±0.012	1.128^{abc} ±0.070	1.816^{a} ±0.130
P2TS1	1.743^{a} ±0.073	1.158^{a} ±0.063	1.138^{ab} ±0.096	1.826^{a} ±0.064	1.189^{a} ±0.066	1.009^{ab} ±0.051	0.556^{ab} ±0.013	1.104^{bcd} ±0.066	2.262^{b} ±0.161
P2TS2	1.715^{a} ±0.079	1.218^{ab} ±0.072	1.221^{a} ±0.086	1.833^{a} ±0.062	1.100^{a} ±0.051	1.092^{b} ±0.049	0.565^{ab} ±0.013	1.118^{bcd} ±0.066	2.240^{b} ±0.125
P2TS3	1.701^{a} ±0.075	1.407^{bc} ±0.079	1.265^{ab} ±0.087	1.866^{a} ±0.050	1.096^{a} ±0.055	0.967^{ab} ±0.052	0.557^{ab} ±0.013	0.969^{ab} ±0.049	1.762^{a} ±0.105
P3TS1	1.718^{a} ±0.050	1.454^{c} ±0.049	1.102^{ab} ±0.063	1.840^{a} ±0.028	1.134^{a} ±0.031	1.004^{ab} ±0.042	0.548^{ab} ±0.007	0.891^{a} ±0.029	2.010^{ab} ±0.116
P3TS2	1.673^{a} ±0.031	1.179^{a} ±0.030	1.087^{ab} ±0.055	1.850^{a} ±0.035	1.130^{a} ±0.038	1.075^{b} ±0.050	0.568^{ab} ±0.008	1.016^{abc} ±0.035	1.830^{a} ±0.174
P3TS3	1.856^{a} ±0.094	1.180^{a} ±0.091	1.153^{ab} ±0.066	1.851^{a} ±0.053	1.117^{a} ±0.041	1.013^{ab} ±0.063	0.558^{b} ±0.015	0.985^{bc} ±0.047	1.941^{ab} ±0.088

From the results of fish length (Table 1) data collected for 60 days is as follows, at 50 ppm immersion for 18 hours (P1TS1) it is 1.6733 ± 0.047cm smaller than 12 hours (P1TS2) and 24 hours (P1TS3) which is 1.946 ±0.063gr and 1.873±0.031cm, while the 60ppm treatment with 12 hour, 18 and 24 hour soaking (P2TS1), (P2TS2) and (P2TS3) had no difference in fish length, namely, 1.866±0.050cm, 1.096±0.055cm and 1.833 ± 0.064 cm, and 70 ppm treatment with 24 hour soaking (P3TS3) is smaller, namely 1.855 ± 0.035 cm compared to 12 hour and 18 hour soaking (P3TS1) and (P3TS2), namely 1.909 ± 0.035 cm and 1.851 ± 0.053 cm can be seen in graph 2. For the Feed conversion ratio (Table 1) in the 50ppm treatment with 12 hours and 18 hours of soaking (P1TS1) and (P1TS2) there was no significant difference, namely 2,114+0.008cm3 and 2,048+0.109cm, while 24 hours of soaking (P1TS3) was greater 1.816+0.130cm. In the 50ppm treatment with 12 hour soaking (P2TS1) it was smaller, namely 2.262+0.161cm, for the 60ppm treatment with 18 hour soaking (P2TS2) and 24 hour soaking (P2TS3) it was 1.830+0.174cm, and 1.941+0.088 cm, whereas in the treatment 70 ppm with 12 hour immersion (P3TS1), namely 2,010+0.116 cm, for 18 and 24 hour immersion (P3TS2) and (P3TS3), namely 1.879+0.013cm, and 1.891+0.013cm can be seen in Figure 4.

Feed conversion ratio (Table 1) from day 30 to day 60 resulted in a difference between treatments of 50ppm, 60ppm, and 70ppm at 12, 18 and 24 hours of immersion. The results of treatment (P1TS3) in 24 hour soaking were 1.843 ± 0.011 compared to (P1TS1) in 12 hour soaking and (P1TS2) in 18 hour soaking, namely 1.666 ± 0.028 and 1.664 ± 0.028, whereas for (P1TS3) in 24 hour soaking the results were 1.888 ± 0.012 greater than (P1TS1) at 12 and 18 hours of immersion (P1TS2) with results of 1.787 ± 0.034 and 1.879 ± 0.013. Likewise, treatment (P1TS3) in 24 hour soaking produced 1.891 ± 0.013, followed by treatment (P1TS2) in 18 hour soaking and treatment (P1TS1) in 12 hour soaking, namely 1.879 ± 0.013 and 1.745 ± 0.035. This can be seen in Figure 5

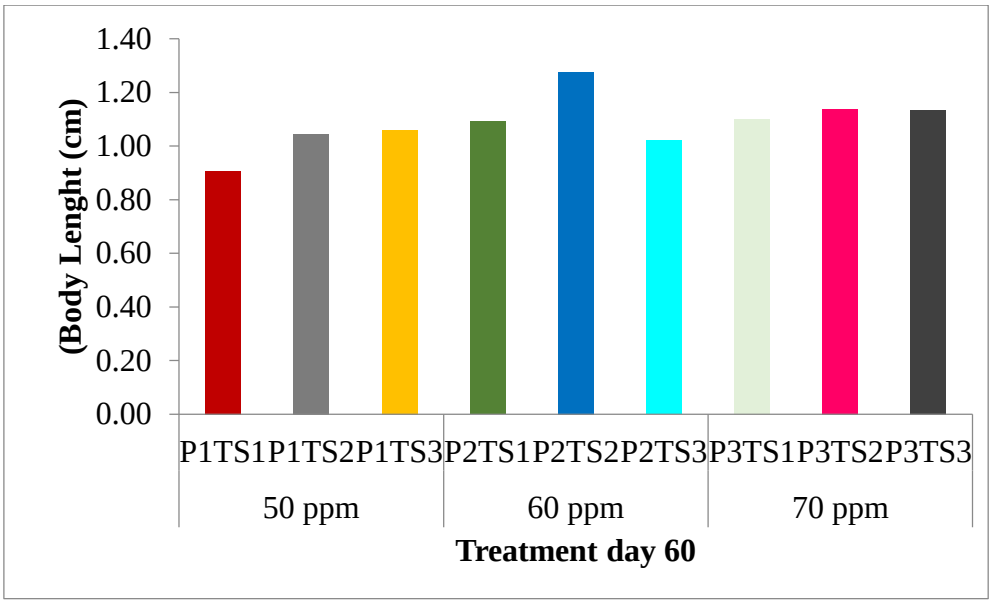
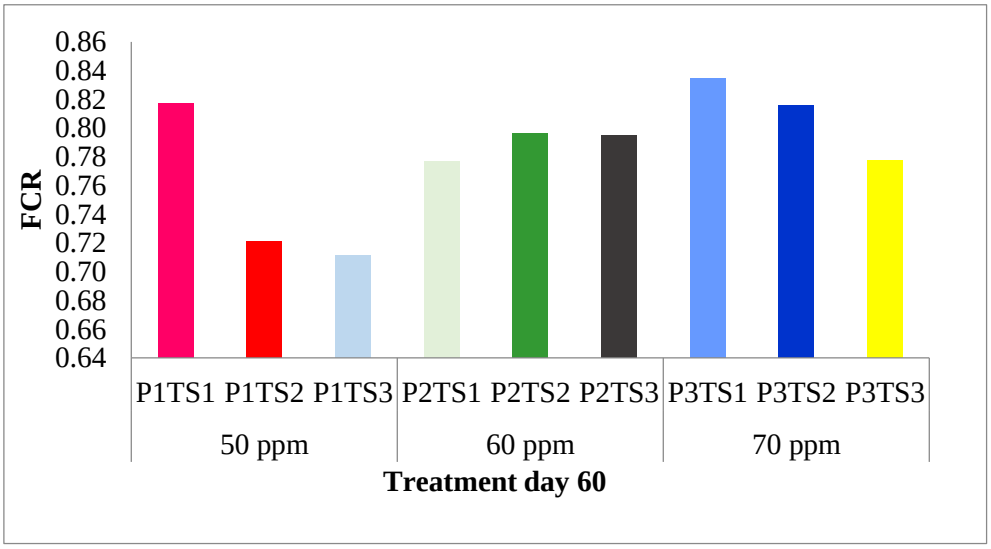


Figure 4. Differences in fish length during the 60 days of research



Gambar 5 Perbedaan FCR pada ikan 60 hari penilitian
Figure 5 Differences of FCR during the 60 days of research

The survival rate results (table 4) show the differences for each treatment from day 10 sMPi to day 60, for treatment P1TS1 the total fish that survived was around 99%, followed by P1TS3 and P3TS3 while treatment P1TS2 was 98% followed by P2TS2, P2TS3 and P3TS1 and in the P3TS2 treatment it was 97% while the lowest was in the P2TS1 treatment at 93%.

In the treatment (P1TS2) with 18 hours of immersion, 29 (99%) fish were alive, while the total number of fish in each treatment was 30 fish. On the 30th day, fish deaths occurred again in the treatment (P1TS2) at 12 hours and 18 hours of immersion, respectively, only 27 (98%) fish remained, followed by the treatment (P2TS1) at 12 hours of immersion, 29 (99%) fish remained alive. fish tail. On the 40th day, dead fish occurred in the

treatment (P3TS1), a total of 29 (99%) living fish. On the 50th day, 28 (98%) fish died in the treatment (P2TS1), and 28 (98%) live fish also occurred in the treatment (P3TS1). On the 60th day of each treatment, the average number of live fish was (98%) for all treatments. This can be seen in Figure 6.

Table 4. The total of Survival rate prom day 10 until day 60 during the study

Treatment	H10	H20	H30	H40	H50	H60	Total
P1TS1	30.00	30.00	30.00	29.00	29.00	29.00	99%
P1TS2	30.00	30.00	30.00	28.00	28.00	28.00	98,5%
P1TS3	30.00	30.00	30.00	29.00	29.00	29.00	99%
P2TS1	30.00	30.00	30.00	26.00	26.00	26.00	93%
P2TS2	30.00	30.00	30.00	28.00	28.00	28.00	98%
P2TS3	30.00	30.00	30.00	28.00	28.00	28.00	98%
P3TS1	30.00	30.00	30.00	28.00	28.00	28.00	98%
P3TS2	30.00	30.00	30.00	27.00	27.00	27.00	97%
P3TS3	30.00	30.00	30.00	29.00	29.00	29.00	99%

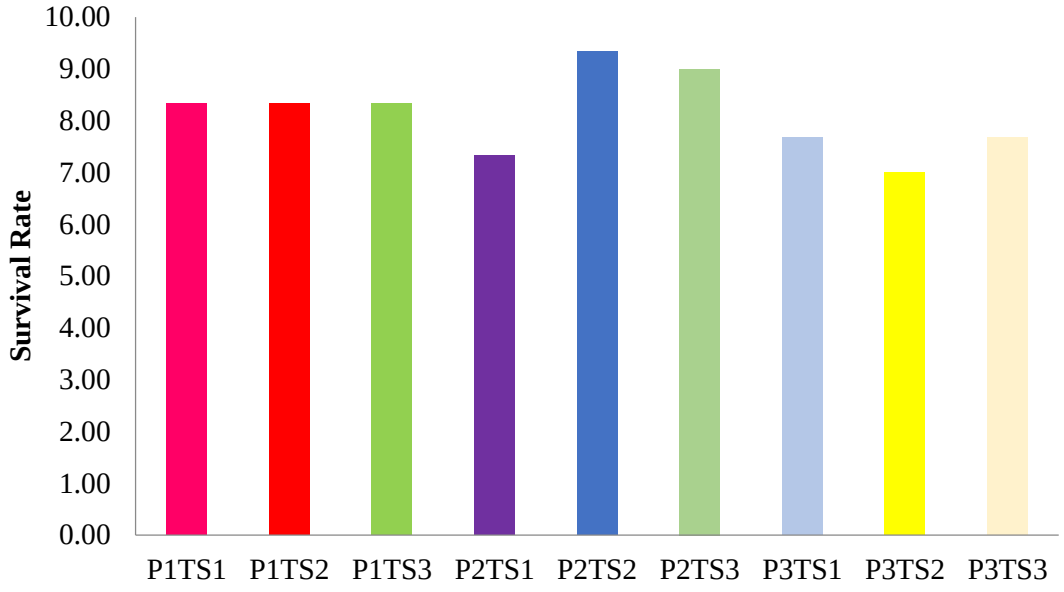


Figure 6. Survival rate with 3 treatment at day 60

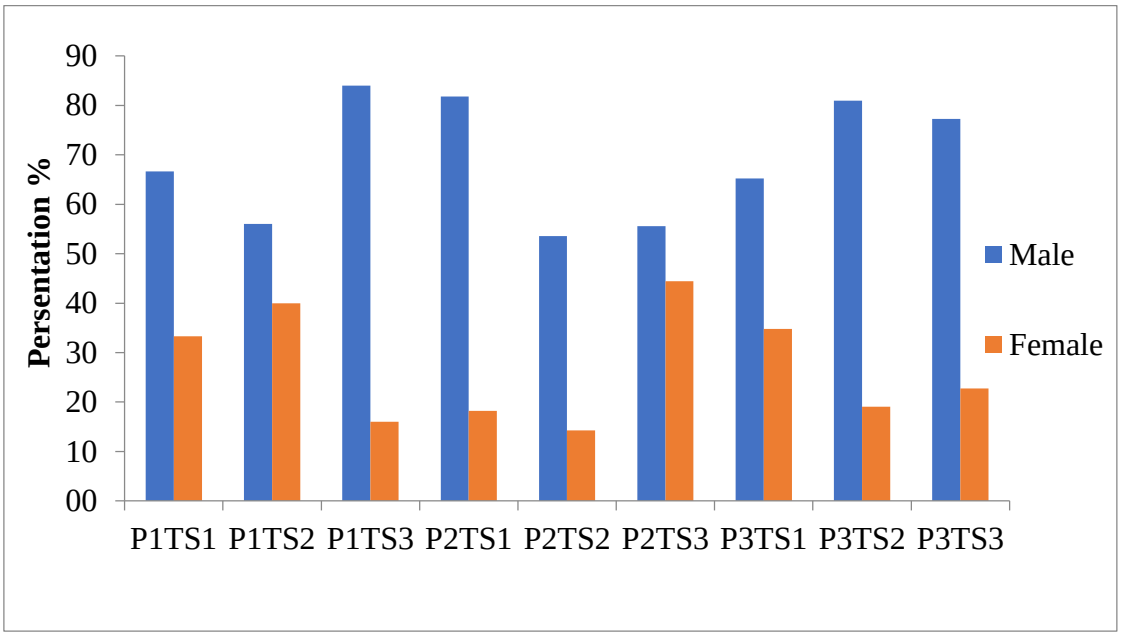


Figure 7. Total percentage of males and females

Based on research results, the male catfish produced in the third treatment produced 80% to 88.9% male fish with the highest value at a concentration of 60 ppm and a soaking time of 18 hours. Where the number of males produced was 172 or 79% and the number of females was 45 or 21%. This can be seen in figure 7.

Discussion

Brahman cattle were developed in the United States, Gulf region, between 1854 and 1926. American Brahmans include Zebu breeds of Kankrey, Ongole, Gir, Krishna, Hariana, and Bhagari. This breed of cattle, which originally developed in the United States, has now spread widely in both tropical and subtropical areas, namely in Australia and also in Indonesia (Sugeng, 1998). The characteristic of Brahman cattle is that they have a large hump and loose skin, wattle under the neck and a wide belly. with many folds (Sakir, 2017). Cow testicle flour is a natural ingredient that is often used in the fish breeding process. Based on several research results using cow testes, it shows that cow testes contain very high levels of the natural testosterone hormone. Apart from that, cow testicle flour is easy to obtain, the price is relatively cheap and the size is large (Hidayani, 2016). Cow testicles contain the natural testosterone hormone which can be used in the masculinization process, namely the effect of changing from female to male. Cow testicles are also easy to obtain, relatively cheap, and large in size. To use it, it needs to be made into flour, so it doesn't rot quickly (E.Yusni et al 2020),

The weight of the fish cannot be measured when the fish is still a larva by giving artemia and tubifex, and when the fish has started to become fry and the fish's body organs are perfect (the fish's mouth has opened) then the fish can be measured by giving pellet type food (E.Yusni et al. , 2020). The application of genital direction techniques is generally carried out on catfish when the larvae are 3-5 days after hatching, the most sensitive time for masculinization (differentiation period). The success of changing sex is not only determined by the type and dose of hormone used, but is also influenced by the length of time the hormone is given, therefore information is needed regarding the optimum length of time for soaking the seeds in a solution of cow testicle flour to change the female sex of catfish to a different type. Male genitals The process of absorbing hormones into the body of Sangkuriang catfish seeds occurs through a diffusion process, from the insan and then through the blood vessels the hormones will flow to the organs and nervous tissue. Arriving at the target cell, the incoming hormone binds to receptors located in the spermatozoa sac (Murni, 2015). Apart from that, by feeding catfish starting on day 30, the weight of the fish begins to increase and the average increase in fish weight is approximately 1 gram/ days as shown in (P3TS2) the weight of the fish weighs 1.856 grams or close to 2 grams per day with 24 hour soaking with a concentration of 70 ppm of cow testicle flour. This is expected to increase if the rearing is carried out in a larger place such as a pond. Meanwhile, this research was carried out in a limited location, namely glass fiber was used. In contrast, the length of the catfish in this study also increased according to (P1TS2), as the weight of the fish increased, the length also increased. 1,851 cm and the increase was also more or less an average of 1 gram/head, namely by giving a fishmeal concentration of 50 ppm with soaking for 18 hours.

The feed conversion ratio (FCR) for catfish in this study also depends on the weight gain of the fish, where the more weight the fish increases, the less feed is needed. In this study, the FCR required on average was almost the same, namely 1.777 gr to 1.946 gr. If linked to theory, this research results in fish body weight and feed requirements being almost the same.

The survival rate for this catfish produces a survival rate of 90% to 93%, including fish that can maintain their life in accordance with previous studies, but depending on the nature and type of fish, as has been studied, 48 hour immersion can only produce 62.41% of Tilapia (*Oreochromis niloticus*) that live (E.Yusni et al 2021). The percentage of cow testicular hormones that can direct sexual differentiation into males (masculinization) is 85%, while females only produce 15% from the treatment of giving 50ppm, 60ppm and 70ppm cow testicle flour and soaking for 12 hours, 18 hours and 24 hours. This is different from research (Irmasari et al., 2012) where giving 3 ml/L of cow testicle flour produced 69.07% male fish.

Conclusion

The effect of giving concentrations of cow testicle flour of 50 ppm, 60 ppm and 70 ppm with 12 hour, 18 hour and 24 hour soaking showed differences. The best fish weight results were in the 50 ppm concentration treatment with 24 hour soaking (P1TS3) and in the 60 ppm concentration treatment with 12 hour soaking (P2TS1) produces males. Meanwhile, the lowest was at 60 ppm with 18 hours of soaking, (P1TS2), the 60 ppm concentration treatment (P2TS3) with 24 hours of soaking and the 60 ppm concentration treatment (P2TS2)

with 18 hours of soaking, and the highest total males were found in the male population, around 79%. 21% female fish.

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

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