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Survival, Growth, Meristic and Morphometric Characteristics of Hybrids Between Black and White Nile Tilapia Strains

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

Tilapia is a commodity that has high economic value. The demand for tilapia continues to increase from year to year. In order to improve the genetic quality of fish, a hybridization program is carried out so that they have advantages becomes superior and good, growth rate, disease resistance and bad environment, and food utilization efficiency compared to their parents. Therefore, this research was conducted at Wanayasa Marine Conservation Service Unit (SPKPD) with A. White Tilapia (♂) x Black Tilapia (♀), B. Black Tilapia (♂) x White Tilapia (♀), C. White Tilapia (♂) x White Tilapia (♀), D. Black Tilapia (♂) x Black Tilapia (♀).This research include the preparation of containers and mains test fish, spawning, fertilization, hatching, maintenance of larvae and fry up to 90 days of age. Then measure the parameters such as survival, growth, meristic and morphometric characteristics. The results of the study showed that the highest specific survival and growth were in treatment B of 92.67%; 19.68%. Morphometric characteristics obtained the highest results in the

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hybrid strain Black Tilapia (♂) x White Tilapia (♀) with a total length of 11.58 cm, head length of 3.21 cm, standard length of 9.32 cm and body height of 3.37 cm. While the highest meristic characteristics were obtained in Treatment B = D.XVI-XVII : 13-14 P. 12-13 V. I : 5 A. III : 9-11 C. II : 15-16 LL. 26-27. This marked with the formula D for dorsal, P for pectoral, V for ventral, A for anal, C for caudal, LL for lateral line. While the Roman numerals indicate the number of hard radii, the Arabic numerals denote the weak radii.

Keywords: Hybrid tilapia; meristic; morphometric; survival rate; black strain; white strain.

1. INTRODUCTION

Tilapia is a commodity that has high economic value. The demand for tilapia continues to increase from year to year. Indonesia is the second largest tilapia producer in the world after China with a total production of 1,100,000 tonnes [1]. Tilapia has several well-known strains in Indonesia, including Genetic Improvement of Farmed Tilapias (GIFT), Bogor Enhanced Tilapias Strain (BEST), Genetically Supermale Indonesian Tilapia (Gesit), Citralada, Hitam 69, Japan for International Cooperation Agency (JICA), National Inland Fish Institute (NIFI), Tilapia Ras Wanayasa (Black), Cangkringan and Red Tilapia Strain Janti (Larasati). The white tilapia strain has not been disseminated to the public [2].

The advantage factor of tilapia is that it is able to convert energy from feed into protein well. The ability to convert feed owned by tilapia for 160 grams of calories consumed can produce 30-40 g of protein and its productivity is quite high. Based on these advantages, in a short time it can produce seeds in large quantities [3].

Tilapia Nirvana has an advantage in its growth speed. Black Tilapia weight growth increased by 45% in the third generation (F3) compared to the first generation [4]. Maintenance from the larvae to the size of the weight of 650 grams per head can be achieved in just six months. White tilapia is one of the strains of tilapia in Indonesia, only information about this fish is still not widely found because it has not been disseminated to the public.

There is a phenomenon in fish cultivators around Wanayasa, Purwakarta Regency, which concludes that "borontok" fish have better growth and survival compared to Nirvana tilapia. This "borontok" fish is the result of cross-breeding trials between Nirvana tilapia and white tilapia conducted by the Wanayasa Regional Water Conservation Service Unit (SPKPD) but has not been published scientifically.

In order to improve the genetic quality of fish, a breeding program is carried out. One of the fish breeding activities is hybridization. Hybridization is one of the genome engineering techniques that can be implemented as a biotechnology application in selection activities. With hybridization, new strains can be produced which advantages have compared to their parents in terms of increased growth speed, survival, and sex ratio, as well as color appearance [5]. Hybridization of tilapia has been widely carried out, such as the F6 pilot tilapia with tilapia which affects seed performance on specific growth rates, feed conversion and survival [6]. Therefore a hybridization program is needed for Nirwana Tilapia and White Tilapia to find out: How is the growth and survival of hybrid fish resulting from crosses of Nirvana tilapia and white tilapia and what are the meristic and morphometric characteristics of hybrid fish resulting from crosses of Nirvana tilapia and white tilapia.

2. METHODOLOGY

The research began in December 2022 by spawning Black Tilapia and White Tilapia fish (*Oreochromis niloticus*) until observations were made in May 2023. The location for the implementation of the activities was carried out at the Northern Region Marine and Fisheries Service Branch (CDKPWU), Regional Water Conservation Service Unit (SPKPD) Wanayasa which is the UPTD belonging to the Department of Maritime Affairs and Fisheries of West Java Province. The method of applying hybridization to tilapia is generally the same as the hybridization method for other freshwater fish, the difference is the combination of hybridization that the male and female organisms wish to carry out. The stages of conducting this research included preparation of containers and test fish broods, spawning, fertilization, hatching, maintenance of larvae and fry up to 90 days of age. Then measure the parameters that will be carried out namely survival, growth, meristic and morphometric characteristics.

2.1 Hybridization Procedure

Hybridization Procedure carried out in several stages based on Rahmawati [7]:

2.1.1 Site selection

The selected location is in the location of the Nirvana tilapia cultivation center, to be precise at the North Region Maritime Affairs and Fisheries Service Branch (CDKPWU), Wanayasa Regional Water Conservation Service Unit (SPKPD). This is to facilitate the provision of fish, feed, ponds, fishing equipment and human resources in carrying out research.

2.1.2 Container installation

The containers in this study were 12 x 2 x 1 x 1 m hapa and 12 x 5 x 2 x 1 m hapa which were installed on the side of the same pond. Hapa size 2 x 1 x 1 m is used for spawning process and hapa size 5 x 2 x 1 m is used for fish rearing process.

2.1.3 Parent selection

The parents used in this study were 6 female tilapia tilapia, 6 male tilapia, 6 female white tilapia and 6 male white tilapia. The criteria for selecting this parent were the condition of the mother being healthy, her body complete and not disabled, aged around 6 months and weighing 250–300 g [8]. This study used broodstock with the first gonad maturity or hens which produced around 130 to 150 broodstock larvae.

2.1.4 Tilapia hybridization

The selected parents were put into a 2 x 1 x 1 m hapa as many as 2 tails (1 male and 1 female). Fish were fed commercial feed in the form of floating pellets (33% crude protein). Feeding is done at satiation 2 times a day. This research was conducted by natural spawning of 12 pairs of Black tilapia and White tilapia broodstock. Thus there are 4 spawning treatments with 3 replications, namely:

A. White Tilapia (F) male (♂) >< Black Tilapia (N) female (♀)
B. Black Tilapia (N) male (♂) >< White Tilapia (F) female (♀)
C. White Tilapia (F) male (♂) >< White Tilapia (F) female (♀)
D. Black Tilapia (N) male (♂) >< Black Tilapia (N) female (♀)

2.1.5 Larvae care

Larvae were produced after 22 days since the broodstock were put together in a 2 x 1 x 1 m hapa. Spawning larvae were taken and put into hapa size 5 x 2 x 1 m each of 100 individuals. During rearing, the larvae are given fish meal.

2.1.6 Fish seed maintenance

Larvae that had been placed in a 5 x 2 x 1 m hapa with a total of 100 individuals were reared and growth observed every 14 days by measuring the weight and length of the individuals reared. The maintenance time is 90 days and given commercial feed with a protein content of 39-41% as much as 5% of the biomass. Feed is given 2 times a day in the morning and evening. Survival observations are carried out every day and removal or separation of dead fish is carried out.

2.2 Observed Parameters

2.2.1 Survival rate calculations

Observation of the degree of survival of fish is carried out every day. Effendi states that to determine the degree of fish survival, the following formula can be used [9]:

$$\mathrm{SR}\ (\%)\ = \frac{Nt}{No} \times 100\%$$

Description :

SR : survival rate during observation
Nt : number of fish at the end of observation
No : number of fish at the start of observation
t : 90 days

2.2.2 Specific Growth Rate / SGR

Growth observations were carried out every 14 days by measuring the weight and length of individual tilapia. Specific growth can be known using the formula [10]:

$$\mathrm{SGR}\ = \frac{\mathrm{Wt} - \mathrm{Wo}}{t} \times 100\%$$

Description :

SGR : Specific Growth Rate (%/days)
Wo : initial fish weight (g)
Wt : final fish weight (g)
t : 90 days

2.2.3 Morphometric character

Tests were carried out on hybrid fish that had reached the age of 90 days. The number of fish measured was 30 for each cross. Measurements made on Black Tilapia release documents included total length, standard length, head length, body height [11]. The total length is the length of the fish measured from the leading edge of the snout to the last tip of the caudal fin. Standard length is the length of the fish measured from the leading edge of the snout to the last end of the spine. Head length is the distance between the leading edge of the head and the rear end of the gill plate. Body height is measured at the highest point between the dorsal and ventral parts, where the part of the base of the fin that crosses the dorsal line is not measured [12].

2.2.4 Meristic character

The meristic character relates to the counting of the number of fins on the body parts of the fish. Tests were carried out on hybrid fish that had reached the age of 90 days. The number of fish measured was 30 for each population of crosses. Measurements made in the Black Tilapia release document included counting the number of dorsal fins, pectoral fins, pelvic fins, anal fins, tail fins and linea lateralis [11].

2.2.5 Heterosis

Heterosis is obtained by comparing the phenotypic values of the variables measured from tilapia parents and their hybrid offspring. This heterosis aims to determine success in developing hybrids from crosses of the parent [13]. The characters that were calculated for heterosis in this study included weight and total length. According to Tave, the value of heterosis can be formulated as follows [14]:

$$H = \frac{\text{Offspring Weight Average} - \text{Parent Weight Average}}{\text{Parent Weight Average}} \times 100\%$$

Description :

H = Heterosis
Offspring Weight Average = Hybrid Tilapia Average
Parent Weight Average = Parent Tilapia Average

2.2.6 Water quality

The quality of a waters is an important requirement that can affect the survival, development, growth and level of fish production. A good environment is necessary for the survival of aquatic organisms. Water quality checks are carried out every 7 days. Several parameters determine water quality, namely temperature, pH and DO [15].

2.3 Analysis Method

Survival and growth data were statistically analyzed using the F test followed by Duncan. In addition, data regarding meristic and morphometric test results were analyzed descriptively quantitatively. The results of water quality testing were compared with water quality standards according to SNI 6141:2009 concerning the production of black tilapia (*Oreochromis niloticus* Bleeker) seed class and the data were analyzed descriptively.

3. RESULTS AND DISCUSSION

3.1 Survival Rate

Survival rate is the percentage of the number of fish that live from the number of fish during the rearing period of 90 days. The results of the percentage of survival rate of tilapia in Fig. 2. shows that the highest percentage of survival rate is found in treatment B of 92.67%. While treatment A was 91.67%, treatment D was 91.33% and finally treatment C was 89.33%. The results of the one way ANOVA statistical test showed that the treatment given had no significant effect ($P > 0.05$) on the survival rate of tilapia.

The high percentage of survival rate obtained shows good results where based on SNI 6141-2009 the survival rate for tilapia aged 90 days is 70%. The results of previous studies on nirvana tilapia had a survival of >90% and white tilapia survival of 93.33% [11]. Hybrid and reciprocal survival obtained above 90% and higher than SNI 6141:2009. Hybrid and reciprocal survival obtained above 90% and higher than SNI 6141:2009. This is in line with research conducted by Rahman, who concluded that hybrid tilapia can improve the genetic quality of tilapia to increase weight and survival of tilapia [16].

3.2 Specific Growth Rate / SGR

Specific growth rate is the percentage growth rate of weight gain per day during the rearing period of 90 days. The percentage results for the specific growth rate of tilapia are shown in Fig. 2.

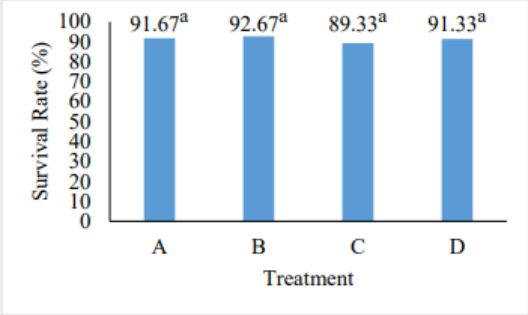

Fig. 1. Graph of tilapia survival rate

Description : A) White Tilapia (♂) x Black Tilapia (♀), B) Black Tilapia (♂) x White Tilapia (♀), C) White Tilapia (♂) x White Tilapia (♀), D) Black Tilapia (♂) x Black Tilapia (♀).

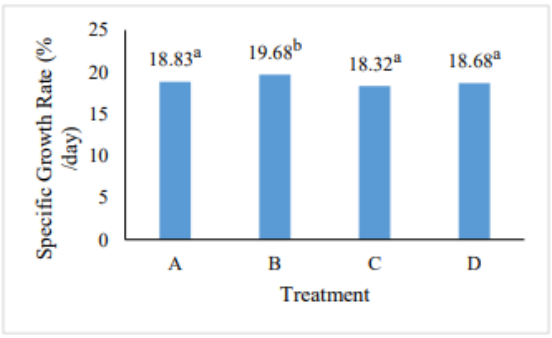

Fig. 2. Graph of specific growth rate tilapia

Description : A) White Tilapia (♂) x Black Tilapia (♀), B) Black Tilapia (♂) x White Tilapia (♀), C) White Tilapia (♂) x White Tilapia (♀), D) Black Tilapia (♂) x Black Tilapia (♀).

The results showed that the percentage of the highest specific growth rate was in treatment B of 19.68%, followed by the percentages of treatments A and D of 18.83% and 18.68%. While the lowest percentage in treatment C was 18.32%. The results of the analysis of variance showed that the spawning treatment of hybrid fish resulting from crosses of Nirvana tilapia and white tilapia had a significant effect ($P<0.05$) on the specific growth rate.

High specific growth in treatment B which is a hybridization of Black Tilapia (♂) x White Tilapia (♀) is in accordance with the hypothesis which results in faster growth. This is presumably due to genetic factors for better growth than the previous parent. This statement is supported by a statement according to Saparinto & Susiana, stating that growth can be influenced by several factors including internal factors which include age, genetics, hormones, the ability to utilize feed and the body's resistance to disease [17].

3.3 Morphometric Character

The morphometric characters measured were total length, standard length, head length, body height as attached in Table 1.

Based on the measurement results in Table 1, treatment B has a longer total length than the other treatments, namely 11.58 cm. Meanwhile, the head length measurement for treatment B was 3.21 cm longer than the other treatments. In the standard length measurement, the highest standard length was obtained in treatment B of 9.32 cm and in the height measurement the results obtained in treatment B were the highest with a length of 3.37 cm.

The results obtained indicated that the morphometric characters of the four treatments did not differ from one another. This is consistent with the results obtained on measuring tilapia in controlled cultivation areas with the same water quality where the values obtained do not differ from one another [18].

Table 1. Observation results of morphometric characters

Treatment	Average ± SD			
	Total length (cm)	Head Length (cm)	Standard Length (cm)	Height (cm)
A	11,52 ± 0,17	3,19 ± 0,07	9,24 ± 0,24	3,36 ± 0,11
B	11,58 ± 0,14	3,21 ± 0,07	9,32 ± 0,26	3,37 ± 0,10
C	11,50 ± 0,19	3,17 ± 0,06	9,18 ± 0,30	3,34 ± 0,10
D	11,51 ± 0,21	3,18 ± 0,07	9,19 ± 0,30	3,35 ± 0,10

Description : A) White Tilapia (♂) x Black Tilapia (♀), B) Black Tilapia (♂) x White Tilapia (♀), C) White Tilapia (♂) x White Tilapia (♀), D) Black Tilapia (♂) x Black Tilapia (♀)

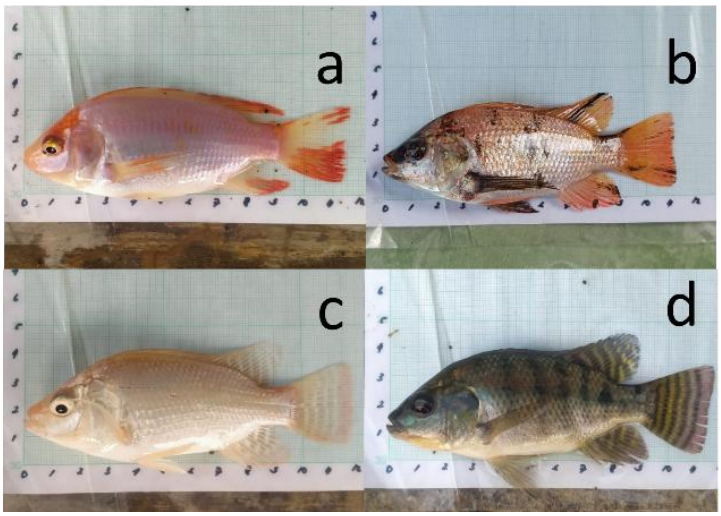

Fig. 3. Observed tilapia

Description : A) White Tilapia (♂) x Black Tilapia (♀), B) Black Tilapia (♂) x White Tilapia (♀), C) White Tilapia (♂) x White Tilapia (♀), D) Black Tilapia (♂) x Black Tilapia (♀)

3.4 Meristic Character

Meristics are characteristics related to the number of certain parts of the fish body that can be used to describe descriptions of fish species, or used to identify unknown species.

Based on the results of color observations of white tilapia A (♂) x Nirvana tilapia treatment tended to be red in color and in treatment B Black Tilapia (♂) x White Tilapia had pale red with black spots and the dominance of the gene controlling the dark color of the parents. This is because white tilapia has been selected and enriched by inserting the red color gene from albino red tilapia as a result of individual selection from Philippine red tilapia [19].

Whereas in treatment C Tilapia Nirvana (♂) x White Tilapia (♀) is dominant in pale white and in treatment D Black Tilapia (♂) x Tilapia Nirvana (♀) is predominantly black and this follows Mendel's law as reported [20]. The pattern of inheritance of colors in crosses between strains in this study also follows Mendel's laws and is influenced by the dominance of dark color control genes.

Table 2. Meristic character observation results

Treatment	Dorsal	Pectoral	Ventral	Anal	Caudal	LL
A	D.XVI-XVII : 11-12	P. 12-13	V. I : 5	A. III : 9-10	C. II : 15-16	LL. 23-24
B	D.XVI-XVII : 13-14	P. 12-13	V. I : 5	A. III : 9-11	C. II : 15-16	LL. 26-27
C	D.XVI : 11-12	P. 12-13	V. I : 5	A. III : 9-10	C. II : 15-16	LL. 23-24
D	D.XVI-XVII : 13-14	P. 12-13	V. I : 5	A. III : 9-11	C. II : 15-16	LL. 26-27

Description : A) White Tilapia (♂) x Black Tilapia (♀), B) Black Tilapia (♂) x White Tilapia (♀), C) White Tilapia (♂) x White Tilapia (♀), D) Black Tilapia (♂) x Black Tilapia (♀)

Based on Table 2, the meristic character calculation results are marked with the formula D for dorsal, P for pectoral, V for ventral, A for anal, C for caudal, LL for lateral line. While the Roman numerals indicate the number of hard radii, the Arabic numerals denote the weak radii. The results showed that the meristic characteristics of the Black Tilapia (♂) x White Tilapia (♀) hybrid strain were the same as the Black Tilapia (♂) x Black Tilapia (♀) hybrid strain.

Saanin, stated that fish generally have the characteristics of the beginning of the dorsal fin above the pectoral fin base, the dorsal fin is longer than the anal fin [21]. Linnea lateralis is a line on the body formed by pores, can be found in fish with scales and without scales. The shape and number of scales that form the lateral line generally varies. Linea lateralis in fish generally has one line, but there are fish that have several. Meristic characters have a genetic basis, but environmental components can modify the expression of these characters during larval development, so that the environment can affect offspring [22].

3.5 Heterosis

Heterosis is obtained by comparing the phenotypic values of the variables as measured by the difference in yield between the first offspring of the results (F1) of the cross with the average of the two parents or the results of the best parents [23]. The calculated heterosis values include the total weight and length contained in Table 3.

Based on Table 3, the heterosis value obtained using the Tave formula shows a positive but low value for the heterosis value for the character of growth in weight and total length. The heterosis value obtained for the weight is 2.176% and the total length is 0.185%. These results indicate that the hybrid tilapia strain Black and white tilapia strain has an advantage of 2.176% of the parental weight, 0.185% of the total length and can be a new genetic source.

According to Robisalmi et al., in the BEST ♀ x Black ♂, the long heterosis value was 4.82% and the weight was -2.22%. The Black ♀ X BEST ♂ has a long heterosis value of -2.48% and a weight of -2.87% [8]. Based on this study, the heterosis value of tilapia hybrids of Nirvana strain and white tilapia strain was higher than previous tilapia hybridization studies and showed dominant offspring.

3.6 Water Quality

The quality of water in rearing tilapia hybrid strains Nirvana and white tilapia was observed for 90 days every 14 days at 06.00, 16.00 and 01.00 WIB.

The results show in Table 4 that the water quality obtained is temperature 21.3-27.1 °C, pH 6.7-7.1 and DO (dissolved oxygen) 3.6-4.3 mg/L. Based on the water quality standards in the Indonesian National Standard (SNI) 6141:2009, the results showed that the temperature and pH were appropriate, but the DO in the observations was lower than 5. According to Pramleonita et al. that the dissolved oxygen content in a tilapia culture medium is at least 4.0 mg/liter, so it can be said that the DO in this research vessel is optimal for tilapia and can support the growth and survival of tilapia [24].

Table 3. Hybrid heterosis values of tilapia (*Oreochromis niloticus*) black strains and white tilapia strains

No.	Character	Offspring Average (gr)	Parent Average (gr)	Heterosis (%)
1	Weight	18,507	18,113	2,176
2	Total length	11,527	11,506	0,185

Table 4. Results of observation of water quality

No.	Water Quality	Results	SNI 6141:2009
1	Water Temperature (°C)	21,3-27,1	25-30
2	pH	6,7-7,1	6,5-8,5
3	DO (dissolved oxygen) (mg/L)	3,6-4,3	>5

4. CONCLUSION

Based on the results of the research conducted, the following conclusions can be drawn:

1. The highest survival was found in the hybrid Black Tilapia strain (♂) x Tilapia White (♀) of 92.67%. While the highest specific growth was found in hybrid fish strain Black Tilapia (♂) x Tilapia White (♀) of 19.68%
2. Morphometric characteristics obtained the highest results in the hybrid Black Tilapia (♂) x White Tilapia (♀) with a total length of 11.58 cm, head length of 3.21 cm, standard length of 9.32 cm and body height of 3.37 cm.
3. Meristic characteristics were found in hybrid fish strain Hybrid fish strain Black Tilapia (♂) x White Tilapia (♀) = D.XVI-XVII : 13-14 P. 12-13 V. I : 5 A. III : 9-11 C. II : 15-16 LL. 26-27. Referring to the discussion that has been presented, it is advisable to carry out further research on this hybrid fish as a new genetic kinship analysis by analyzing hybrid tilapia black strain and white tilapia strains to determine its kinship to other fish.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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