



Phenotypic diversity in three generations of domesticated Asian redbtail catfish, *Hemibagrus nemurus* (Valenciennes, 1840) in Indonesia

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Abstract. It is necessary to characterize the phenotypes of each produced generation to evaluate the rate of changes to support the success of a domestication program. This study aimed to analyze the phenotype diversity of three domesticated Asian redbtail catfish (*Hemibagrus nemurus*) generations (G-1, G-2 and G-3) based on parameters such as truss morphometry, growth performance, and survival rate. The truss morphometric analysis was performed using 30 individuals sampled from each generation. Growth performance and survival rate analysis were experimentally carried out using a completely randomized design (CDR) with three replications. Rearing of the three Asian redbtail catfish generations began with an acclimatization period for one week and continued through up to 90 days. Rearing was done in 9 concrete ponds (3 x 2 x 1 m) with a circulation system. The water level in each pond was maintained at 80 cm. The stocking density was 100 individuals m⁻³. Fish were fed on commercial pellet containing protein (28%) at a 5% feeding rate (of the total biomass of fish). The results of truss morphometric analysis canonical function on 21 characteristics showed differences in characters occurring in the midsection (B3) and rear body of the fish (C1, C3, and C5). The analysis of intrapopulation similarity index revealed the highest values in G-1 and G-2 (60.0 %), whereas the lowest was observed in G-3 (40.0%). The results of domesticating three generations of Asian redbtail catfish for 90 days showed the G-3 had the highest length growth performance, SGR, FCR and survival rate ($p < 0.05$) which were 6.49 ± 0.39 cm; $0.70 \pm 0.03\%$ per day; 2.3 and 67.67%, respectively.

Key Words: Asian redbtail catfish, phenotype, morphometric truss, growth, generations.

Introduction. The Asian redbtail catfish (*Hemibagrus nemurus*) is an important economic freshwater fish. Indeed, it is considered as one of the 22 freshwater genetic resources in Indonesia that can be used to diversify the business of aquaculture (Gustiano et al 2015). So far, the demand of the Asian redbtail catfish for both consumption and aquaculture activities still depends on the wild catch. The redbtail catfish farming has been going on in Indonesia since 1991 (Gaffar & Muflikhah 1992). However, its production has not yet reached its optimum, and there are still issues such as reproductive performance to be managed. The productivity of redbtail catfish is considered to be low. Subagja et al (2015) reported that the survival rate of the redbtail catfish at the size of 5 cm is only about 10-30%. Meanwhile, Radona et al (2018) stated that the survival rate of the redbtail catfish reared indoor at room temperature (25-26°C) is around 50%.

Consequently, the low productivity of the Asian redbtail catfish can lead to a lack of fish availability, resulting in the unfulfillment of the demands of both aquaculture activities and consumption. Thus, efforts are required to improve the productivity of Asian redbtail catfish, one of which is through domestication programs. Domestication is a process of changes in living organisms involving genetic changes that occur from generation to generation and can control the reproductive cycle and growth (Mylonas et al 2010; Lorenzen et al 2012; Teletchea & Fontaine 2014). Proper broodstock

management and larvae rearing are crucial in domestication activities (Teletchean & Fontaine 2011; Bijaksana 2012).

Mapping the genetic diversity of a population is required in supporting the success of domestication programs. Genetic diversity is a key parameter for the fitness population to ensure the sustainability of a community. According to Dunham (2004), the genetic diversity was associated with the ability to adapt to environmental changes. It can be identified based on the measurement of the morphological characteristic of the phenotype following the morphometrics and growth (Sneath 1995). Phenotype characterization can evaluate the rate of changes in each produced generation (Bagherian & Rahmani 2009). The present study aimed to analyze the phenotype diversities in three generations (G-1, G-2, and G-3) from the domestication of the Asian redtail catfish based on truss morphometric, growth performance and survival rate.

Material and Method. This study was conducted at Germplasm Research Station which is an installation unit of the Institute for Freshwater Aquaculture Research and Fisheries Extension (BRPBATPP) Bogor, in July-October 2017.

Fish sample. Fish, used as the sample, came from the domesticated Asian redtail catfish seeds of three generations. The predecessor of the Asian redtail catfish originated from Cirata Lake, Cianjur district, West Java. The formed population of each generation was obtained through mass spawning at a ratio of 1:2 (1 male to fertilize two females) from the previous generations. The broodstock was selected based on rapid growth parameter. Spawning of broodstock in the G-0, G-1, and G-3 was conducted through the induction of gonadal maturity using HCG 500 IU kg⁻¹ (spawning was carried out 24 hours post-injection). LHRH analog was injected at doses 0.6 mL kg⁻¹ for female and 0.2 mL kg⁻¹ for the male to stimulate ovulation. The first generation of Asian redtail catfish (G-1) had a standard length (SL) of 7.78±0.41 cm and a total body weight (BW) of 7.39±0.96 g. The second generation (G-2) had a SL of 8.28±0.21 cm and a BW of 8.06±1.10 g, while the third generation (G-3) had a SL of 7.34±0.11 cm and a BW 6.47±0.39 g.

Morphometric observation. Morphometric was conducted on 30 individual samples randomly taken from each generation (G-1, G-2, and G-3). Truss morphometric was evaluated through measurement and determination methods of truss point based on Radona et al (2017a, b). The 21 measurable characters were as follows: A1 (end of mouth -latter part of skull), A2 (end of mouth - the lower end of operculum), A3 (end of mouth -beginning of first ventral fin), A4 (latter part of skull - the lower end of operculum), A5 (the lower end of operculum - beginning of first ventral fin), A6 (latter part of skull -beginning of first ventral fin), B1 (latter part of skull - beginning of first dorsal fin), B3 (latter part of skull - beginning of anal fin), B4 (beginning of first dorsal fin - beginning of anal fin), B5 (beginning of first ventral fin - beginning of anal fin), B6 (beginning of first dorsal fin - beginning of anal fin), C1 (beginning of first dorsal fin - beginning of second dorsal fin), C3 (beginning of first dorsal fin - end of second dorsal fin), C4 (beginning of first dorsal fin - beginning of anal fin), C5 (beginning of anal fin - end of anal fin), C6 (beginning of first dorsal fin - end of anal fin), D1 (beginning of first dorsal fin - beginning of upper part of caudal fin), D3 (beginning of first dorsal fin - beginning of lower part of caudal fin), D4 (beginning of upper part of caudal fin - end of anal fin), D5 (end of anal fin - beginning of lower part of caudal fin) and D6 (beginning of upper part of caudal fin -end of lower part of caudal fin). The truss points were made by putting the fish on the paper sheet coated with clear plastic and placed within a styrofoam. Each point was marked with a needle following the pattern in truss morphometry (Figure 1). The results of the truss morphometry measurement of all the characters were converted into a ratio by dividing the value of the character measured by the value of the SL.

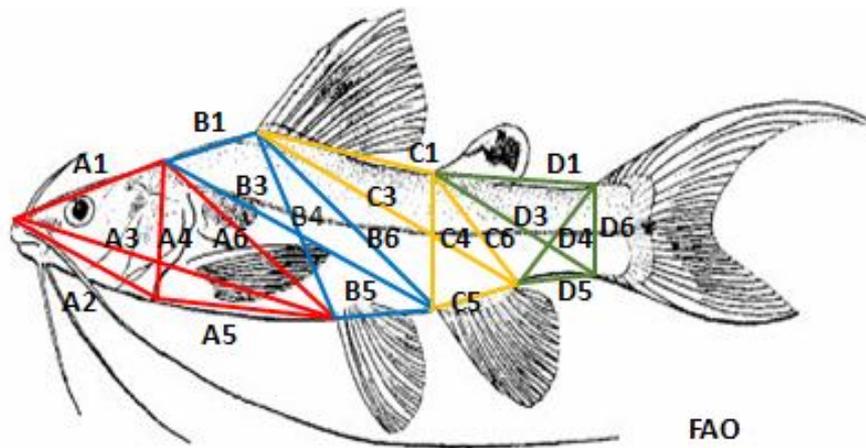


Figure 1. Measurement points in truss morphometry on Asian redtail catfish.

Growth performance. The observed growth parameters were total length gain (cm), total weight (g), specific growth rate (SGR, %), survival rate and feed conversion ratio (FCR). This study was experimentally conducted using a completely randomized design (CDR) with three replications. The rearing of the three generations of Asian redtail catfish begun with an acclimatization period (seeds) for one week in the pond and continued for 90 days. The rearing was done in 9 units of concrete ponds (3 x 2 x 1 m) with a circulation system and a water height of 80 cm. The stocking density was 100 individual m^{-3} . During that period, a commercial pellet containing protein (28%) was provided at a ratio of 5% of the total fish biomass. Feeding was carried out three times a day, at 07.00 am, 12.00 pm, and 17.00 pm. The observations of growth and survival rate were done every 30 days. The growth parameters were based on both the length and weight of individuals by taking 30% of the total population per ponds. Fish weight measurements were done by using digital scales (model Notebook series 500 g, 0.01 g) with an accuracy level of 0.01 g. Fish length measurements were conducted using a ruler with an accuracy level of 0.5 mm while the survival rate was calculated based on the number of dead fishes at the end of the experiment divided by the number of fishes at the beginning. FCR measurements were performed at the end of the study following Fry et al (2018) formula.

Analysis of data. Truss morphometric characters were quantitatively analyzed using the ImageJ program. Calculation value of the distribution characters of truss morphometric (intra- and interpopulation) and sharing component index was conducted with ANOVA using SPSS version 18 and presented in the form of diagram canonical function. Data of growth, survival rate and FCR were tabulated using Microsoft Excel 2010, then analyzed by analysis of variance (ANOVA) using SPSS 22.0 followed by a Duncan test at 95% level of confidence.

Results and Discussion

Truss morphometry of three Asian redtail catfish generations. The average values of 21 truss morphometry characters of three Asian redtail catfish generations (G-1, G-2 and G-3) originated from domestication are presented in Table 1. The morphometric diversity was expressed in the coefficient of variation (CV) presented in Table 2.

Phenotype diversities, based on 21 morphometric characters and analyzed using Wilks lambda, showed that only four characters had different values ($p < 0.05$). Whereas, Lavene test analysis (ANOVA) revealed that all measured characters had similarities ($p > 0.05$) (Table 2). The measured characteristics showed different characters in the midsection and rear body of the fish. In the midsection, it occurred in the latter part of the skull - beginning of anal fin (B3), while in the rear body, it occurred at the beginning of the first dorsal fin - beginning of second dorsal fin (C1), beginning of

first dorsal fin - end of second dorsal fin (C3), and beginning of anal fin - end of anal fin (C5).

Table 1

The average value of phenotype diversity of the 21 characters in truss morphometric analysis of three generations (G-1, G-2, and G-3) of domesticated Asian redtail catfish

<i>Truss area</i>	<i>Morphometric characters</i>	<i>Average</i>		
		<i>G-1</i>	<i>G-2</i>	<i>G-3</i>
Head of fish	A1	0.365±0.019	0.360±0.015	0.366±0.016
	A2	0.210±0.019	0.208±0.016	0.203±0.015
	A3	0.255±0.016	0.252±0.014	0.258±0.016
	A4	0.358±0.014	0.352±0.016	0.353±0.012
	A5	0.561±0.012	0.553±0.018	0.554±0.019
	A6	0.176±0.016	0.178±0.014	0.179±0.016
Midsection of fish	B1	0.191±0.015	0.192±0.009	0.193±0.015
	B3	0.170±0.017	0.156±0.017	0.165±0.015
	B4	0.384±0.013	0.391±0.012	0.390±0.013
	B5	0.515±0.029	0.515±0.013	0.518±0.017
	B6	0.261±0.016	0.264±0.014	0.261±0.090
	C1	0.130±0.009	0.142±0.008	0.131±0.011
Rear body of fish	C3	0.332±0.014	0.342±0.012	0.334±0.015
	C4	0.184±0.009	0.187±0.008	0.184±0.006
	C5	0.480±0.014	0.494±0.011	0.487±0.016
	C6	0.173±0.011	0.173±0.008	0.176±0.009
	D1	0.137±0.017	0.132±0.017	0.138±0.022
	D3	0.262±0.018	0.263±0.026	0.268±0.017
Caudal peduncle of fish	D4	0.131±0.006	0.136±0.013	0.132±0.008
	D5	0.300±0.016	0.297±0.025	0.300±0.019
	D6	0.180±0.015	0.177±0.010	0.185±0.016

Table 2

Wilks lambda analysis and Lavene test (ANOVA) coefficient of variation of the 21 truss morphometric characters of three generations of domesticated Asian redtail catfish

<i>Morphometric characters</i>	<i>The coefficient of variation (%)</i>			<i>Significant Wilks lambda</i>	<i>Significant ANOVA</i>
	<i>G-1</i>	<i>G-2</i>	<i>G-3</i>		
A1	5.21	4.18	4.24	0.268	0.198
A2	8.92	7.70	7.43	0.242	0.338
A3	6.39	5.43	6.26	0.351	0.652
A4	3.83	4.48	3.48	0.213	0.574
A5	2.18	3.21	3.37	0.089	0.195
A6	9.38	7.93	8.88	0.731	0.742
B1	8.03	4.79	7.64	0.903	0.108
B3	10.07	10.67	8.99	0.003*	0.994
B4	3.33	3.03	3.30	0.077	0.939
B5	5.63	2.46	3.32	0.780	0.429
B6	5.99	5.48	3.44	0.629	0.052
C1	7.28	5.77	8.47	0.000*	0.400
C3	4.25	3.46	4.39	0.011*	0.348
C4	4.98	4.14	3.41	0.181	0.367
C5	2.90	2.15	3.36	0.001*	0.087
C6	6.43	4.48	4.84	0.413	0.210
D1	12.36	13.15	15.64	0.483	0.158
D3	6.94	9.99	6.28	0.487	0.897
D4	4.72	9.65	5.74	0.184	0.159
D5	5.26	8.45	6.47	0.840	0.532
D6	8.11	5.87	8.67	0.100	0.053

notes: * significantly different ($p < 0.05$).

The coefficient value of character diversity indicated the level of variability of the corresponding character in a population. The level of a character phenotypic variability reflects the population genotypic variability (Falconer & Mackay 1996; Gjedrem et al 2012). The coefficients of variation (CV) of the Asian redtail catfish population originating from domestication (G-1, G-2, and G-3) ranged from 2.15 to 15.64%. The low value of the CV in the Asian redtail catfish from domestication was similar to those of other native species such as tinfoil barb (Kusmini et al 2016) and giant gourami (Radona et al 2017b). A significant test of Wilks lambda and ANOVA were conducted to determine the character that can be used as a characteristic of a fish. The result of canonical function analysis (Figure 2) showed that individual morphological characters of the Asian redtail catfish population (G-1, G-2, and G-3) interconnected with one another and spread evenly across all quadrants (1, 2, 3 and 4).

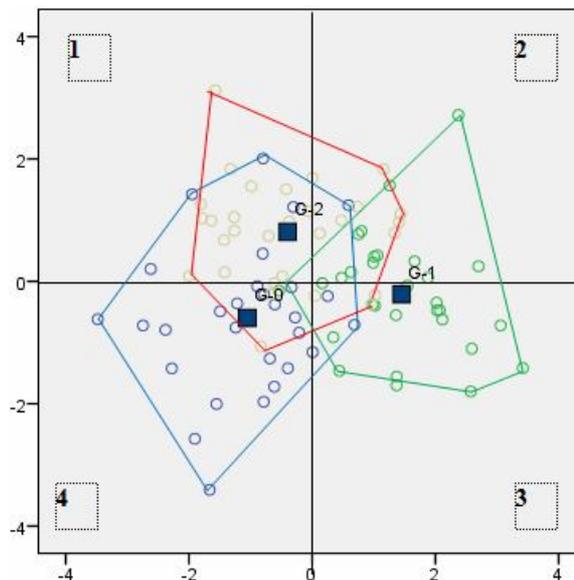


Figure 2. Individual morphological character distribution in a population consisting of three generations of Asian redtail catfish from domestication.

Based on the analysis, the sharing component index intrapopulation phenotypes indicated that the Asian redtail catfish in G-1 and the G-2 population had the highest sharing component index, which was 60.0%, compared to the G-3 population with 40.0% (Table 3). The lowest sharing component index of interpopulation was observed in G-1 and G-2 (10.0-13.3%) and G-1 and G3 (26.7-30.0%); G-2 and G-3 (30.0%).

Table 3
The percentages of sharing component of the three Asian Redtail catfish generation originating from domestication

<i>Population</i>	<i>G-1</i>	<i>G-2</i>	<i>G-3</i>	<i>Total</i>
G-1	60.0	13.3	26.7	100.0
G-2	10.0	60.0	30.0	100.0
G-3	30.0	30.0	40.0	100.0

In the domestication process, intrapopulation sharing component index value obtained in the Asian redtail catfish was lower than that of the tinfoil barb. Radona et al (2017a) reported that the value of intrapopulation sharing component index in the domestication of the tinfoil barb from West Kalimantan was 76.7%. The low value of the intrapopulation sharing component index in the Asian redtail catfish was a consequence of the high genetic diversity that occurred as a result of the domestication process. Mahardika et al (2011) stated that the higher the value of the intra-interpopulation sharing component,

the lower the genetic diversity would be. Meanwhile, lower value of intrapopulation sharing component resulted in higher genetic variability. The phenotype diversity is a key parameter of the population fitness that ensures its sustainability and passive response to natural or artificial selection (Lorenzen et al 2012). Phenotype diversity can give an idea of the fitness status of the population. The higher the diversity of the phenotype, the higher the ability of fish to adapt to environmental changes will be.

Meanwhile, low diversity of the phenotype can decrease both adaptability and productivity. The low diversity of the phenotype is due to the availability of the effective number of breeding broodstock in the aquaculture process which is very little, thus, increasing the potential of inbreeding (Gjedrem et al 2012). According to Ujjania & Kohli (2011), the phenotype of a fish is generally indicated by a different potential for growth. In relation with the domestication effort, one aspect that has to be considered and play an important role in the domestication program is the provision of broodstock for breeding to increase the productivity of the Asian redtail catfish.

Growth performance of the three generations of Asian redtail catfish originating from domestication.

The results of rearing three generations of Asian redtail catfish from domestication during 90 days showed that the G-3 had optimal growth performance with a standard length of 6.49 ± 0.39 cm and a specific growth rate of $0.70 \pm 0.03\%$ per day. Statistically, those values of the length growth were significantly different ($p < 0.05$) compared to those of the G-2. Also, the FCR (2.3) of G-3 revealed significant differences when compared to that ($p < 0.05$) of G-1 with 2.5.

The FCR shows the utilization of nutrients in the feed by the fish. Thus, a low FCR value indicates that the use of the feed is efficient. Feed efficiency is the proportion of fish biomass that increases along with the amount of feed consumed (Luo et al 2015; Xia et al 2017). High feed efficiency obtained in the G-3 demonstrated a better feed utilization compared to G-2 and G-1. The growth parameters such as length, weight, specific growth rate and FCR of the three generations of Asian redtail catfish from domestication are presented in Table 4.

Table 4
Length gain, weight gain, specific growth rate, and FCR of Asian redtail catfish from domestication

<i>Growth parameters</i>	<i>Population</i>		
	<i>G-1</i>	<i>G-2</i>	<i>G-3</i>
Initial length (cm)	7.78 ± 0.41	8.28 ± 0.21	7.34 ± 0.11
Initial body weight (g)	7.39 ± 0.96	8.06 ± 1.10	6.47 ± 0.39
Final length (cm)	14.21 ± 0.16	13.33 ± 0.23	13.83 ± 0.23
Final body weight (g)	52.93 ± 1.13	55.48 ± 0.47	55.32 ± 0.42
Absolute length (cm)	6.42 ± 0.54^b	5.04 ± 0.32^a	6.49 ± 0.31^b
Absolute weight (g)	45.54 ± 1.99^a	47.43 ± 1.21^a	48.85 ± 0.37^a
Specific growth rate of length (% day ⁻¹)	0.67 ± 0.07^b	0.53 ± 0.03^a	0.70 ± 0.03^b
Specific growth rate of weight (% day ⁻¹)	2.20 ± 0.17^a	2.15 ± 0.16^a	2.39 ± 0.06^a
FCR	2.5^b	2.1^a	2.3^a

Note: Numbers followed by different superscript letters in the same row indicate significant differences according to Duncan test ($p < 0.05$).

Based on the results of observations during the study, the G-3 Asian redtail catfish had optimal growth performances. Indeed, the G-3 group had a weight increase of 7% compared to G-1, while the length increased about 29% compared to the G-2 group. In general, the domesticated fish had better growth performance compared to wild fish. Growth is a genotype expression in a phenotype that is quantitatively observed and influenced by environmental factors. The increase in growth in each generation is in the form of change/improvement of the genetic quality which is a positive response to environmental conditions (Millot et al 2011; Kusmini et al 2013). Millot et al (2010) stated that domesticated fish tend to have a high appetite that can accelerate the growth

process. The SGR of weight and length were directly obtained and proportional to weight gain and length. The higher the growth increment, the greater the SGR value will be.

The survival rate of the three generations of domesticated Asian redbtail catfish. The survival rate of the three generations of domesticated Asian redbtail catfish reared in concrete ponds for 90 days is presented in Figure 3.

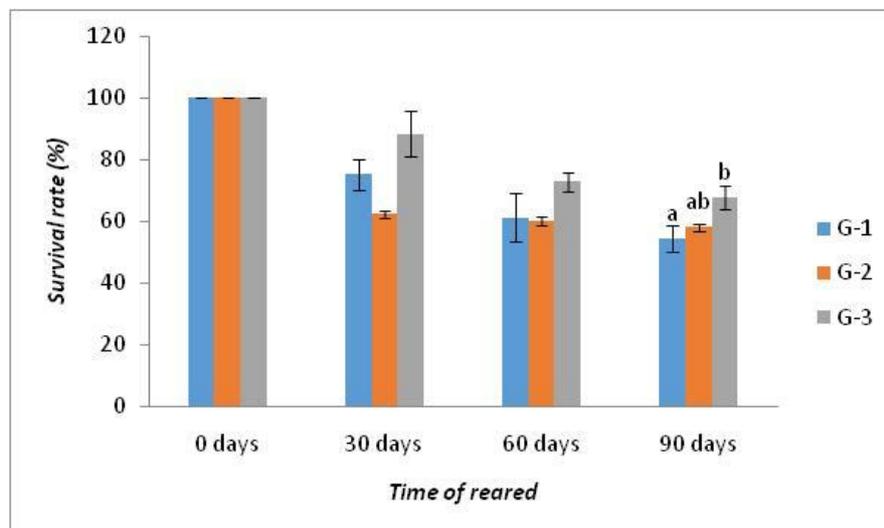


Figure 3. The survival rate of domesticated Asian redbtail catfish.

At the end of the rearing period, the highest survival rate was observed in the G-3 about 67.67%. Statistically, the survival rate of the G-3 population was significantly different ($p < 0.05$) compared to that of the G-1 population, i.e., 54.33%. The high survival rate is presumably due to the G-3 undergoing a suitable process of adaptation in a controlled environment. The success of the domestication program will impact on improving the genetic diversity of fish which is closely related to economic characteristics such as survival and efficiency of feed utilization (Lorenzen et al 2012; Gjedrem et al 2012). Studies have reported that the population of fish in the last generation had a better adaptive performance with the highest survival rate compared to the population of previous generations in fish species such as lalawak fish (Prakoso et al 2017), common carp of rajadanu strain (Radona & Asih 2012).

Conclusions. The morphological characters of the three generations (G-1, G-2, and G-3) of domesticated Asian redbtail catfish revealed differences that occurred in the midsection (B3) and rear body of fish (C1, C3 and C5) with an intrapopulation phenotype sharing component index of 60%. The G-3 had an optimum performance with increased weight (7%), length (29%), survival rate (67.67%) and FCR (2.3).

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