

Gene Pool Of The Body Banding Patterns Of Barbus Tetrasona At North Sumatra

Deny Supriharti, Elimasni

ABSTRACT: Study of gene frequencies and genotype of Sumatra Barbus was conducted to determine the the gene pool of body banding patterns of Sumatra Barbus (*Barbus tetrasona*) This research were done by examined the genotype and gene frequency in its population at North Sumatra. Sample of *Barbus tetrasona* were collected among its distributors at Medan and kept at several fish tanks. The acclimatizations were done for several months to growth and mature. When the fish reached its maturity, male and female were separated in a special fish tanks and induced by using a reproductive hormone (ovaprim 0.5 mg / kg body) for 2 times with an interval 8 hours. Then the male and females of the fish were conducted to mate. After spawning happened, the fry would be separated. The fries were fed by rotifers and maintained until they reach maturity and ready to be mated. The hybrids would be carried out between individuals (P), F1 and F2, also testcross and back cross. Data obtained from those hybrids were analysed by Chi-Square based on Mendel law assumption. The result of the research showed that the majority of *Barbus* crossing were fit with Mendel's assumption, except 2 crossing showed unfitness from Mendel's assumption. The genotype and gene frequencies of the body banding pattern of *Barbus* also in line with Mendel's law. The additive complete dominant trait has the highest genotype and gene frequencies which are 0.62 and 0.55 respectively, and the recessive trait also showed the lowest genotype and gene frequencies, 0.09 and 0.29 respectively. Meanwhile the incomplete body banding had the genotype and gene frequencies 0.29 and 0.16 respectively.

Keyword: *Barbus tetrasona*, gene frequency and genotype frequency

Introduction

Sumatra Barbus fish (*Barbus tetrasona*)

Barbus Sumatra is often called barbus tiger because of its lines pattern on his body (Lesmana and Daetami 2012), At the beginning, this fish were included in *Capoeta tetrasona*. Since 1855, an expert, Bleeker, described that this fish is better called *Barbus tetrasona*. Then Bleeker change the name into *C. Sumatras* in 1860, finally in 1963, Alfred (1963) named this fish into *Barbus tetrasona* after the rectification of the errors form nomenclature. Zakaria - Ismail (1993) said that the naming issue this fish there were frequently confuse in fish taxonomy books, even today when looking for about *Barbus* tiger hence the name that appears is *Barbodes*, *Capoeta* and *Puntius* (in Arifin, 1999) Indonesia is a country that has large biodiversity including different types of fish in the sea and/or freshwater. Among these fishes, many of them have a potential economic including the Sumatran fish *Barbus*. *Barbus* also known as tiger (*Barbus tetrasona*). The main characteristic of this fish is lie on its lines pattern and its color. Majority of these fishes have four vertical lines throughout the body from the eye to the tail, but some has incomplete lines in the the middle part of its body (Lesmana and Daetami 2012) *Barbus tetrasona* known as *Barbus* Sumatran because it comes from rivers in Sumatra but it can also be found in other areas of southeast asia. These fishes move very aggressively and easily to be stress. This fish usually live in small group. Tiger *Barbus*'s body is slightly lean with brownish yellow and silvery. It has four vertical lines which cross into the body and it has black triangular shape of back down fins. The males have red dorsal fin, which light up during spawning time (Lesmana and Daetami, 2012).

The differences between the male and female will be known when the fish is coming at maturity age or size of approximately 5 cm. The males usually have a more slender body with reddish fins while the females has larger and slightly rounded body and they do not have reddish fins (Ferreti, 2015). The differences of its body banding could have survival function. This line patterns, especially those from tropical Cyprinidae group usually associate with protection mechanisms. For example Characidae fish use the line to protect from predators. Therefore, when there is a change of the linease pattern change, it means fish protection behavior is also changing. Thus the observation of the inheritance pattern of lines on a group of fish is very interesting to do (Frankel 2002)

The frequency of the gene (allele) and genotype

In studying the phenotype or phenotype variation of fish, also environmental factors are often ignored. Genotype factors will not work alone without considering environmental factors. From various studies in molecular genetics, the study of how the expression of genetic occurs in phenotype fish were conducted, but the molecular researchers often ignore environmental conditions or habitat of fish. molecular genetics and biotechnology scientists often discuss in detail the research molecular procedure molecular yet rarely pay attention to the environmental aspects of how fish bred so that environmental factors in gene expression are often completely overlooked (Penman, 2000). Population genetics is the study of genotype and allele frequencies in a population based on the ratio of Mendel's laws. By comparing the frequency of allele of species with Null character models, it can be also known whether the evolution on a character happened or not or whether the environment was stable or unstable. Genes will be passed down from one generation the next generation without changes. Each individual will has alleles derived from its parent so that their genotypes or allele frequencies change can be described by Hardy- Weinberg. When the same allele of a group of individuals of the same species are grouped together, it can be formed "gene pool". Allele frequency is only obtained from a population while

- Deny Supriharti, Elimasni
- Biology Department, University of North Sumatra

individuals will not have the allele frequency (Yamaha et al, 2007). Genotype frequency determined by the frequency of allele and also the nature of the characters inheritance from the parent to the child and family, whether homozygot, heterozygous, or irregularities Mendel's laws. So that, the genotype frequency can also be regarded as the abundance of each genotype in a population. (Ramos et al 2012). Allele frequency can also be interpreted as the relative frequency of an allele within a gene locus. By knowing the relative allele frequencies of the fish, it also can describe the behavior of the fish. Genetic population itself can be defined by the study of distribution and changes in allele frequency in a population (Dunhan, 2012). The frequency of genotypes showed genotype did the best at/ or slightly found in a species population. Peroni (2009), Population genetics not only consist of groups of species but a marriage groups in which genes will be transferred from one parent to the child. Similar genes are not related only to an individual but rather how the gene scaled to the offspring as well as the possibility of missing genes or the appearance of new gene. The emergence of such an existing gene in the population must be constantly on the transfer to the next generations but not so with genotype (Pandian, 2000). This aims of the study are to know the frequency of the gene (allele) of body banding patterns of *Barbus tetrazona* as well as its genotype frequency of body banding patterns.

RESEARCH DESIGN AND METHODS

Collecting of samples

Barbus fishes were purchased from various distributors randomized. The fish will be acclimatized on 20L aquariums at a water temperature of about 24 ° C, pH 6.5. The fish were reared into adulthood with a size of approximately 5-7 cm. Adult fishes needed to separate between male and female fish. The male fish is marked with a red ventral fins and slimmer and brighter while females have a rounded body without reddish ventral fins, the body color also slightly paled (Purdon, 1993) Spawning aquarium set up 24

hours before the time of spawning by providing a smooth nylon fabric at the bottom and given a high oxygenation for 24 hours. After that, the female and male fishes moved into the new aquarium. The newly male inserted one hour after spawning female entered in the aquarium. The mating process continues until the female fish fatigue, characterized by less active move. The mating ready when the female fish ready to release all its egg for approximately 2-3 hours. Then the fish is separated from its egg. The egg will hatch after approximately 36 hours. This fish larva needs to be maintained well. Fish larvae will swim freely for 5 days and adequately fed with a small amount but the condition of the oxygen and the water temperature should be maintained properly (Ferreti 2012). To obtain the gene frequencies and genotype frequencies, it was required a lot of the spawning process. Hybridization process must be done between P x P, F1 x F1, F2 x F2 do a back cross between F1 x F2. and also carried out hybridization between *Barbus* that have characters in common at least among the fishes. To accelerate the induction of spawning male and female parent will also be done using reproductive hormones or hormone hipofisis. Then all the offsprings were all puppies will be separated on different aquarium and in news feed rotifers and allowed to mature to be ready crossed again in accordance with the type of crossing is planned. Morphology, especially the pattern of fishing line should be observed and recorded. Phenotype data obtained will be analyzed by chi-square analysis to assuming the pattern of inheritance of genes under the laws of Mendel (Frankel 2002)

RESULTS AND DISCUSSION

Tiger barbus exhibits three phenotypes associated with banding body. One posses four vertical lines throughout entire body, other posses incomplete lines body especially at belly part and the last posses half banded body banding cross at the abdominal of the fish. The result of crossing between parents and their offspring can be seen at the table 1 below.

Table 1: Results of a cross, the ratio of the probability, and the Chi-Square (χ^2) of *Barbus tetrazona*

| No | Parental | | | | Phenotype | | | ratio | χ^2 | |
|----|----------|----------|---|----------|-----------|----|---|-------|-----------|--------------------------|
| | ♀ | genotype | | genotype | ♂ | X | Y | | | Z |
| 1 | P1 | AABB | X | AABB | PI | 5 | 0 | 0 | 1 : 0 | |
| 2 | P2 | AABB | X | AABB | PII | 6 | 0 | 0 | 1 : 0 | |
| 3 | P3 | AABB | X | AABB | PIII | 4 | 0 | 0 | 1 : 0 | |
| 4 | P4 | AABB | X | AABB | PIV | 6 | 0 | 0 | 1 : 0 | |
| 5 | P5 | AABB | X | AABB | PV | 5 | 0 | 0 | 1 : 0 | |
| | | | | Total | | 26 | 0 | 0 | | |
| 6 | P1 | AABB | X | aabb | K1 | 5 | 0 | 0 | 1 : 0 | |
| 7 | P2 | AABB | X | aabb | K2 | 5 | 0 | 0 | 1 : 0 | |
| 8 | P3 | AABB | X | aabb | K3 | 6 | 0 | 0 | 1 : 0 | |
| 9 | P4 | AABB | X | aabb | K4 | 7 | 0 | 0 | 1 : 0 | |
| 10 | P5 | AABB | X | aabb | K5 | 7 | 0 | 0 | 1 : 0 | |
| | | | | | | 30 | 0 | 0 | | |
| 11 | K1 | aabb | X | aabb | KI | 0 | 0 | 3 | 0 : 1 | |
| 12 | K2 | aabb | X | aabb | KII | 0 | 0 | 2 | 0 : 1 | |
| 13 | K3 | aabb | X | aabb | KIII | 0 | 0 | 3 | 0 : 1 | |
| 14 | K4 | aabb | X | aabb | KIV | 0 | 0 | 2 | 0 : 1 | |
| 15 | K5 | aabb | X | aabb | KV | 0 | 0 | 3 | 0 : 1 | |
| | | | | | | 0 | 0 | 13 | | |
| 16 | FIP1K1 | AaBb | X | aabb | KI | 3 | 6 | 1 | 1 : 1 : 1 | 0.1000;0.2000; 0.9000 |

| | | | | | | | | | | |
|----|--------|-------|---|------|------|-----|----|----|-----------|--------------------------|
| 17 | F1P2K2 | AaBb | X | aabb | KII | 8 | 10 | 1 | 1 : 1 : 1 | 2.2237;0.0263; 2.9605 |
| 18 | F1P3K3 | AaBb | X | aabb | KIII | 9 | 6 | 0 | 1 : 1 : 1 | 7.3500;0.3000; 3.7500 |
| 19 | F1P3K4 | AaBb | X | aabb | KIV | 9 | 6 | 0 | 1 : 1 : 1 | 7.3500;0.3000; 3.7500 |
| 20 | F1P5K5 | AaBb | X | aabb | KV | 6 | 6 | 0 | 1 : 1 : 1 | 3.0000;0.0000; 3.0000 |
| | | | | | | 33 | 34 | 2 | | |
| 21 | KK11 | aabb | X | AABB | P1 | 5 | 0 | 0 | 1 : 0 | |
| 22 | KK21 | aabb | X | AABB | P2 | 4 | 0 | 0 | 1 : 0 | |
| 23 | KK31 | aabb | X | AABB | P3 | 6 | 0 | 0 | 1 : 0 | |
| 24 | KK41 | aabb | X | AABB | P4 | 4 | 0 | 0 | 1 : 0 | |
| 25 | KK51 | aabb | X | AABB | P5 | 6 | 0 | 0 | 1 : 0 | |
| | | | | | | 26 | 0 | 0 | | |
| 26 | FF11 | AaBb | X | AaBb | F11 | 8 | 8 | 1 | 9 : 7 : 1 | 0.2546;0.0422; 3.3962 |
| 27 | Ff12 | AaBb | X | AaBb | F12 | 6 | 7 | 0 | 9 : 7 : 1 | 0.2348;0.3016; 0.8125 |
| 28 | Ff13 | AaBb | X | AaBb | F13 | 7 | 8 | 0 | 9 : 7 : 1 | 0.2457;0.3112; 0.9375 |
| 29 | Ff14 | AaBb | X | AaBb | F14 | 7 | 8 | 2 | 9 : 7 : 1 | 0.6855;0.0422; 0.8335 |
| 30 | Ff15 | AaBb | X | AaBb | F15 | 10 | 6 | 3 | 9 : 7 : 1 | 0.0445;0.6451; 5.5301 |
| | | | | | | 38 | 37 | 6 | | |
| | | Total | | | | 153 | 71 | 21 | 245 | |

X: complete pattern of barbuis; Y: incomplete pattern of barbuis; Z: half banded

The probability for all test is ≥ 0.05 , $df = 2$ is 5.99. Therefore among all the test, only two test is not fit to the expected ratio according to Mendel's assumption. Table 1 showed the data of Barbuis crossing. The number of offspring observed from the crossing clearly showed the genotypes of body banding at Barbuis was fit to mendel's assumption that stated by Frankel (1985). The body banding patterns are controlled by two autosomal gene loci acting additively, with complete dominance at each locus. Among those mating only 2 crossing that did not fit to the Mendel's assumption, the others clearly present fitness to the Mendel's assumption. The unfitness of these two crossing could be as a result of excessive inbreeding among the population. Barbuis tetrazona is a fish that already well known and well spread as ornamental fish. Majority fish

distributors get the fish from crossing each other. It is rarely taken from the wild population already. Eventhough Barbuis Sumatra were originally come from Sumatra, but it is uneasy to find the fish at wild population anymore. Therefore, the more this fish domesticates, the more unstable the population become. Thus, it could affect the gene pool of the genotypes and its gene of the body banding pattern of Barbuis tetrazona. This condition, if happens at such long periode, it could lead into evolution of the gene of the body banding pattern of Barbuis. The inheritance of body lines pattern are interesting to be learn. Generally, the emergence of the differences of the body lines pattern related to protection mechanism. It is probably raise an evolution such a long time ago especially from tropical cyprinids as shown in characidae (Frankel, 2002).

Table 2 showed the gene pool of Barbuis tetrazona at North Sumatra at this present.

Table 2: The gene pool of Barbuis tetrazona

| | | | |
|-----------|--------|-------------|----------|
| Frequency | aabb/r | aaB /A_bb/q | A_B /p |
| Genotype | 0.09 | 0.29 | 0.62 |
| Gene | 0.29 | q = 0.16 | p = 0.55 |

The data is served as initial data of the gene pool of body banding pattern of Barbuis tetrazona. There was abundant researches of Barbuis tetrazona were already conducted. But, none of data presented related to genetic quantitative. Therefore, this research tried to fulfilled the lack data, especially from Barbuis tetrazona at North Sumatra. According to Dunham (2012), the gene existency and originality of the characters will hard to determine when there is no genetic population data. Eventhough, there are 2 crossing of Barbuis tetrazona showed unfitness to Mendel's assumption, the majority crossing gave results fit to mendel's assumption. This result was consistent with their genotype and gene frequencies. Based on the total number of offspring from all the crossing done, the

genotype and gene frequencies of body banding patterns of Barbuis are in line with Mendel's assumption. It clearly showed that the dominant trait has the highest genotype and gene frequencies which are 0,62 and 0.55 respectively, and the recessive trait also showed the lowest genotype and gene frequencies, 0.09 and 0.29 respectively. In the future, this initial data can be compared into other quantitative data of the similar trait of the fish Drastic changes in allele and genotype frequencies showed significant changes in habitat or behavior of fish (Dunhan, 2012).

CONCLUSION:

Majority crossing of Gene pool research of body banding patterns of *Barbus tetrazona* were fitted into Mendel's Assumption which is controlled by two autosomal gene loci acting additively as proposed by Frankel (1985). From the data obtained, it can be deduced that dominance trait had the highest genotype and gene frequencies which are 0.62 and 0.55, and the recessive trait also showed the lowest genotype and gene frequencies, 0.09 and 0.29 respectively

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