The Relationship of Body Length and Ratio Pappilla with Sex in Gobi Fish (\textit{Sicyopterus macrostetholepis})

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Abstract
Research about the relationship of body length and ratio papilla with sex in gobies (\textit{S. macrostetholepis} Blkr.) has been done at Animal Structure and Developmental Laboratory, Biologi Department, Faculty of Mathematics and Natural Sciences, Andalas University, Padang, which purposed to analyse the relationship of body length and ratio papilla with sex of gobies (\textit{S. macrostetholepis} Blkr.). The samples were taken in wild stream area at Batangkurani river, Padang City. This research used descriptive method and data were analyzed by qualitatively and quantitatively. The results of investigation showed that in several gobies (\textit{S. macrostetholepis} Blkr.) with different sex had the same of body length and the same of ratio papilla. So, there was not relationship between of body length and ratio papilla with sex. Goby fishes (\textit{S. macrostetholepis} Blkr.) it belongs to the hermaphrodite protogini, which the androgynous young females, while in adulthood, it would change sex to male. The results of this study are expected to add to the treasures of knowledge and information about reproductive gobies (\textit{S. macrostetholepis} Blkr.) in the preservation and development of fish farming.

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INTRODUCTION

Fish are members of cold-blooded vertebrates (poikilothermik) which live in the water, moving with fins, the body has scales and breathing with gills. The fish has a number of species diversities with more than 27,000 in the world. The fish can be found in all of he puddles like in freshwater, brackish water or salt water at variation depth. The fish also has a high economic value either as a food ingredient or as a pet. In Indonesia, most of the utilization of fisheries is still rooted in fishing from nature. The arrests were carried out continuously causes decreasing of fish population.

Decreasing of fish population caused by fishing activities which are not controlled, it can lead to overfishing and the destruction of habitat, thus, it will effect the environment and the existence of biological activity in these waters, which could threaten the sustainability of the source itself. It is estimated that the rare fish in Indonesia is increasing, with the increasing of illegal fishing activities and exploitation being offset by conservation activities. Therefore, to maintain fish populations still in high population and sustainable, it needs cultivation. In cultivation, Biology aspect of fish reproduction is very important. At least, it consist three types of fish reproduction. One of them is hermaphrodite. According to (Effendi, 2002); (VICTOR, 2010) Elacatinus rubigenis, is described from Utila in the Bay Islands of the Gulf of Hon-duras (Western Atlantic) a hermaphrodite is an interesting phenomenon in fish reproduction. An individual is said to be a hermaphrodite fish when the body is having ovarian tissue as a determinant of individual females and testicular tissue as a determinant of individual males. One group of fish that includes hermaphroditic fish is from the family Gobiidae.

Gobies are the largest tribe widespread fish in the waters of temperate and tropical region (ZAMRONI, 2016); (LARSON et al., 2016). In addition, these fish are also a lot of mangrove forests in encounter. (Fahrian et al., 2015) states that found 6 glodok fish in magroveat Mororejo village, Kendal Regency. These fishes can be found in the waters of salty, brackish and freshwater, as well as public waters such as lakes and rivers. According (Devi, 2012), (LARSON et al., 2016) a special feature of this fish has pelvic fins together and a from of suction disc which allows them to remain in position in the fast-flowing waters. Small, but thick with thinning tail shape, sirip- thick fins by having two dorsal fins. According (VICTOR et al., 2010); (VICTOR, 2010) Elacatinus rubigenis, is described from Utila in the Bay Islands of the Gulf of Hon-duras (Western Atlantic, some kind of family Gobiidae, it belongs to the hermaphrodite protogoni, which the androgynous young females, while in adulthood, it would change sex to male.

To distinguish males and females can be seen from the primary and secondary sexual characteristics. Primary sexual characteristics of fish is characterized by the organ that is directly related to the reproduction process that ovarian veins and vessels for females and testes with veins and vessels for the male fish. Secondary sexual characteristics, they can be seen on signs of the primary sexual characteristics, such as shape, color or other organs (PERISTIWALDY, 2006). By Ghufron (2005), based on the length of the fish’s body supposed can be determined by gender differences in fish. To distinguish the sex of male and female fish, it can be done by observing the physical form of the body fish with body length sightings. If the physical characteristics can not be done, the gender distinction can be made by observing the fish genital organs (papilla).

Research on the papilla observations have been made by Burhanuddin & Genisa (1984), the gobies Periophtalmodon schlosseri and Boleophthal musboddarti live in the waters of the estuary of the Banyuasin (Palembang). The male fish, its papilla is in long form, while the female fish, its papilla is spherical shape. But the gobies S. Macrostetholepis Blkr that live in the waters of Batangkuranji river, Padang City, particularly in females who undergo sex change, until now there is no information about it. Therefore, research on the relationship of the body length and the papilla with sex ratio needs to be done in order to add information about the reproductive biology of gobies, as well as assist in developing the breeding gobies programs.

From the description above, it can be formulated that problems which need to be answered from this study. It is how long the relationship of the body and papilla ratio gobies (S. macrostetholepis Blkr.). The purpose of this study was to determine the relationship of the body length and the papilla ratio gobies (S. macrostetholepis Blkr.) with sex. The results of this study are expected to add to the treasures of knowledge and information about reproductive gobies (S. macrostetholepis Blkr.) in the preservation and development of fish farming.
METHODS

This research was descriptive method. Body length measurements and observations of the ratio (length and width) papilla are associated with gonads histological structure examination, as obtained in sampling. The tool is used in this study the stereo zoom binocular with motic image plus program, and the light microscope with a magnification of 40X100. A set of surgical tools, petridishes, tissue, paper labels, bottles films, ring case for embedding, lights spritus, Microtome American Optical with a thickness of 8 μm, the dyebath for 16 units, glass objects, glass covers, the incubator temperature to 50° C, Caliper varnir with a precision of 0.01 mm. Fishing gear centrums (Electric Snatcher) and stationery. The materials used papilla and gonad gobies. Physiological solution, bouin, alcohol series 70-100%, paraffin hard, aquadest, xilol, hematoxylin, eosin, poly l-lysine, and entelan.

Sampling was carried out while gobies in their reproductive years. The samples were obtained by 196 tails using fishing gear centrum (Electric Snatcher) on the fast-flowing areas in the waters of Batangkuranji river, Padang City. Geographically located at 0° 48' – 0° 56' and 100° 21' LS-100° 33' BT, with a long stream of approximately 17 km with area 22.149.32 ha (Bapedalda Padang City, 2004). The measured parameters such as length and width papilla which starts from the edge of the anus until the hole papilla (Rodgers, 2005). Measurements papilla used Stereo zoom binoculars. Stereo zoom binoculars are equipped with a camera and connected directly to the computer so that the image can be directly extracted and measured. The program used software Motic Image Plus.

Preparation of histological gonadin order to know the sex of gobies. Gonad gobies that have taken the data from it morphometrik, then it is isolated for histology preparations which made semi-sheer follow Paraffin method. The gonads were washed with physiological solution, fixed with fixative solution (Bouin) for 24 hours, then dehydrated in alcohol series 70-100% for 1 hour, purification with xilol for 1 hour, infiltrated with paraffin and planted on the beam cutter. Then, it sliced crosswise using a microtome (American Optical) with pasting on the slide, and continued thickness of approximately 8 μm with the coloring process haematoksilin eosin staining and deparafinisasi with xilol.

Mixture is examined under a light microscope with a magnification of 4x100 to see its histology structure. From observation, the sex cells are found, the epitelial layer, connective tissue, gonad developmental stages to find out the sex. Furthermore, the fish are grouped by gender. Subsequent histological is preparations which represents photographed.

To determine the relationship between body length and ratio papilla S. macrostetholepis Blkr., it is analyzed using simple linear regression, with the formula Sudjana (2016) as follows: Simple Linear Regression on formula: \( Y = a \pm bX \)

RESULTS AND DISCUSSION

Characteristics of S. macrostetholepis Blkr.

Number of S. Macrostetholepis Blkr. caught from Batangkuranji river, Padang that are used for sample are 196 tails. Having observed their morphological characteristics, they could be divided into males and females since S. Macrostetholepis Blkr. is dimorphism. According to (Victor et al., 2010); (Victor, 2010) Elacatinus rubrigenis, is described from Utila in the Bay Islands of the Gulf of Hon-duras (Western Atlantic; (Rodgers, 2005), sexual dimorphism is a morphological characteristic that can be used to distinguish males and females.

Characteristics of morphological S. macrostetholepis Blkr. female individually are fat and slow. It makes slow in swimming. In the body, there is a black stripe look like ribbons which numbering 4-8 pairs, and a dark brown body color, (Figure 1a). Meanwhile, in the male individual body has color brighter than the females, which is mauve with streamers 6-8 line pairs and caudal fin has orange color which is surrounded by a black line on the rim (Figure 1b). According Agromedia (2002), the male fish brightly colored than the female fish.

Lestrel (2000) states the male is smaller and slimmer than the females which is elliptical. From the results, the male is smaller than the female fish. This body shape is necessary in order to move swiftly, especially while doing the activity. These fishes move in the mud by immersing pectoral fins simultaneously and push it backward when his straight forward and rigid. Elongated dorsal fin has an orange thorn, when breathing the throat of young male fish has an orange color (Peristiwaldy, 2006).

From observations that have been made, the shape of the papilla of the female is generally elongated with a small base section towards the widened end and rounded as split in half like the fans (Figure 2a), while in males, it elongated widened towards the end and a small rounded (Figure 2b). Burhanuddin & Genisa (1984), stating
that the shape of male fish papilla are elongated and rounded end portions like an inverted triangle, while the female papilla elongated shape with two split ends. According to (Victor et al., 2010); (Victor, 2010) Elacatinus rubrigenis, is described from Utila in the Bay Islands of the Gulf of Honduras (Western Atlantic, papilla in males fish is as genital and papilla in female fish is as the form of holes which serve as an estuary of urine and sperm or eggs.

**Figure 1.** Photo of *S. macraostetholepis*. A = Gobi fish female. B = Gobi fish male

**Relationship with Body Length Ratio papillae**

The measurement results refers to the observations of the characteristics *S. macrostetholepis* Blkr. by observing the characteristics of fish, it is obtained 131 female and 65 male fish tail. It is Based on measurements of body length and papilla ratio of female fish and male fish that live in the waters of Batangkuranji river, Padang City, it can be seen in Table 1.

Based on Table 1, it can be seen, the average body length of the female 59.18 ± 9.06 mm, whereas in males 56.93 ± 9.01 mm. Papilla ratio of females has an average of 1.83 ± 0.36 and 0.81 ± 0.15 for male fish. From the above data, female body length has greater than the length of the male body. This condition also occurs in papilla ratio, the ratio of female papilla is larger than the males. Hutomo & Naamin (1982), states that in gobies (*Periophthalmus koelreuteri*) males are smaller than females. To see the connection and the length of fish body papilla ratio, it is done by using simple linear regression. The test results can be seen in Figures 3 and 4.

**Table 1.** The Average body length and ratio papilla *S. macrostetholepis* Blkr.

<table>
<thead>
<tr>
<th>Measurement</th>
<th>Body Size Length (mm)</th>
<th>Ratio papillae</th>
</tr>
</thead>
<tbody>
<tr>
<td>Females</td>
<td>Males</td>
<td>Females</td>
</tr>
<tr>
<td>Average</td>
<td>59.18 ± 9.06</td>
<td>56.93 ± 9.01</td>
</tr>
</tbody>
</table>

The relationship between body length and the ratio of the papilla on *S. macrostetholepis* Blkr. can be shown in the equation $Y = 2.0643 - 0.0039X$ with $r = 0.098$ and it is readable, each increasing in body length 2.0643 units, the papilla ratio decreased 0.0039 units for female fish. In the male fish, body length each increased 0.5388 units, the papilla ratio increased 0.0048 units, the equation $Y = 0.5388 + 0.0048X$ and $r = 0.248$. Results showed that there was no effect on the body length ratio of the papilla, which increase the body length in females and males. It did not occur accretion papilla ratio. Although in some
the length of the body male fish, it also showed the increase of the ratio of the papilla. Genital papilla fish is a bulge erectile act as a conduit of sperm or eggs (Burhanuddin & Genissa, 1984). Then from the observations that have been made, there are some fish of different sexes having a body length range and papillae same ratio. Gender on the fish has nothing to do with the length of the body and the ratio of the papilla.

Figure 3. Regression graph of body length and Papilla ratio female fish

Figure 4. Regression Graph of body length and papilla ratio male fish.

The length of the body in fish is influenced by various factors, physical factors (temperature), chemical factors (dissolved oxygen and acidity of water) and biological factors (amount and type of food) (Kusmawati, 2009). Ghufron, (2005) states that in nature the existence of genetic variation in the growth potential of fish for the same type on different populations, they are usually influenced by environmental factors such as temperature and food availability.

The Analysis of Histology gonads S. macrostetholepis Blkr.

To strengthen the data about gender differences based on the length of the body and the ratio of the papilla on S. macrostetholepis Blkr. Then observation of histological structure of fish gonad are needed to be done. The measurement data were then grouped based on histologic observations of the gonads. The histologic observations showed that there was different stages of gonad development in females, adult females and intersex females of 131 females consisting of adult females of 83 and 48 females intersex.

Adult females

In observation of histological preparations gonads, mature female fish showed that it almost has the same structure in all samples that observed, where the tunica albugenia is thin which built by serous layer. In the medulla area, it looks a lamella-lamella. They are drawn up by the oocyte. Oocytes dominated tertiary relatively equal, while the primary oocytes are not so many, and interstitial tissue is narrow. This is in accordance with the opinion of Suherman (2001) that the gonads are in a stage of development characterized large-sized oocytes. The increasing size of the oocyte cause interstitial tissue narrowing.

Agency yolk (vitellogen) already meets some of the oocyte. It is found some irregular nucleus. It indicates the condition of the oocyte in a state of degeneration (atresia). According to Suherman (2001) that the formation of the yolk (vitellogenesis) oocytes can degenerate.

Figure 5. Histology gonad S. macrostetholepis Blkr. adult females. Magnification of 10 x 100. Ta = Tunica albugenia, It = interstitial tissue, L = lumen, Op = primary oocytes, In = irregular nucleus.

Female Intersex

Histological observations of gonad female fish stocks intersex are showed at different stages of development, but they show further structures. It is marked by the presence of sperm and the seminiferous tubules. It looks interstitial tissue thickening and lumen clearly visible. In the cortex, the tunica albugenia is visible thickening and fibrous connective tissue with collagen fibers are arranged very dense, which continue to enter between the oocyte. As a result, there is formation of cavities between the lamella and ovaries and oo-
cytes. Some oocytes are degenerated. Suherman (2001) states that the degeneration of oocytes began in the core area, in the central part of the core are material (unknown). It is enabled splitting into two which become four, four become eight seemed to occupy all parts of the oocyte.

To see the connection body length of the fish and the fish papilla ratio of adult females and female intersex, it do simple linear regression. The test results in Figure 8 and 9.

### Table 2. The Average length of the body and the ratio of fish papilla adult females and female intersex.

<table>
<thead>
<tr>
<th>Measurement</th>
<th>Body Size Length (mm)</th>
<th>Ratio papillae</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Adult female</td>
<td>Female intersex</td>
</tr>
<tr>
<td>Average</td>
<td>59.83 ± 9.21</td>
<td>58.06 ± 8.79</td>
</tr>
<tr>
<td>Stdv</td>
<td>5.92</td>
<td>8.79</td>
</tr>
</tbody>
</table>

Figure 7. Histology gonad *S. macrostetholepis* Blkr. Male. Magnification of 4 x 100. Spd = spermatid, L = lumen, Sp = Spermatocyte, Ta = Tunica albugenia, Tbs = Seminiferous tubules, Sp = Spermatocyte, L = lumen, Oa = Degenerate of oocyte, It = interstitial tissue.

Male

At this stage of gonad, the development of male fish are difficult to observe because the network is very small (Figure 7). According Fujaya (2004), at the rate of testicular development is dominated by primary spermatocytes. In preparation histology male gonads, it can be seen tubular semineferus which surrounding the sperm. It has been in the lumen. While tubular semineferus, there are spermatids at different stages. Spermatids in inside the tubules semineferus will undergo metamorphosis (without undergoing the cell division). It develops into spermatozoa that are functional.

Relations Body Length and Ratio papillae On Adult Females Fish and Fish female intersex.

Histological observation results showed the differences in the female gonad and developmental stages. It is namely adult females and female intersex. Based on observations of the length, the body and the ratio of the papilla are on the second phase of the data obtained which shown in.

From the table above, it can be seen that the average adult female body length is greater than the females intersex where the average adult female body length is 9.21 ± 59.83 mm, while females intersex 58.06 ± 8.79 mm. Something similar, it happened on the ratio of the papilla, where the average adult female ratio is 1.85 ± 0.37 and 0.34 ± 1.79 female intersex.

From the graph, it can be seen that the body length has a high correlation with the ratio of the papilla for both adult females and female intersex. The equation for each is as follows:

- **Adult Female Fish**: $y = 21.25 - 0.004x$, $r^2 = 0.112$
- **Female Intersex**: $y = 2.016x - 0.0099x$, $r^2 = 0.308$

Figure 8. Graph of body length regression and Papilla ratio adult female fish.

Figure 9. The visible size of the papilla ratio remains despite a growing body length.
Every increasing in body length by 2.1254 units, the papilla ratio decreased by 0.0045, where the regression equation $Y = 2.1254 - 0.0045X$ and $r = 0.110$. As for the female fish intersex, the regression equation is $Y = 2.0165 - 0.0039X$ and $r = 0.098$. From these results, it illustrates that the length of the body did not affect the ratio of the papilla, which increase the body length in adult female and female fish intersex. It did not happen accretion papilla ratio. A sex change in fish had nothing to do with the length of the body and the ratio of the papilla.

A sex change on *S. macrostetholepis* Blkr. does not occur in a particular body length, as well as the ratio papilla. A sex change from female to male in *S. Macrostetholepis* Blkr., it believed that occur spontaneously. It is influenced by various factors. One of the factors that can cause changes gender in a social factor. According to Rodgers (2005), the social group of fish in the wild, a male came to power peak charge a minimum of seven fish females. Protagonist hermaphrodite fish is in the colony, as males leave the colony then the tail of the female fish will turn out to be male and to the top.

Gobi fish is one type of the hermaphrodite protogyny fish. Hermaphrodite protandre is a group of hermaphrodite fish, which in one life cycle, there is a process of gonad differentiation from male to female phase. (Victor, 2010) Elacatins rubrigenus, is described from Utila in the Bay Islands of the Gulf of Honduras (Western Atlantic, priyono, has explained that based on the development of ovarian tissue and testes present in an individual will determine the kinds of hermaphroditism, synchronous, protogyny and protandre. Hermaphrodite synchronous is a group of fish which in one life cycle consisting female sex cells and male sex cells that can fuse together. (Ratna & Abdulgani, 2012) hermaphroditic protogyny is a group of hermaphrodite fish which in one life cycle poses a process of gonad differentiation from the female phase to the male. While Hermaphrodite protandre is a group of hermaphrodite fish that in one life cycle poses a process of gonad differentiation from male to female phase.

Hermaphrodite protandre and hermaphroditic protogyny are often called as consecutive hermaphrodite. It is when young fish have gonads that are organized into two kinds of sex, where there are tissue testes and ovaries in one of them that has not been developed well (Effendi, 2002). According to (Victor et al., 2010), in succession or other terms known as “Sex Reversal” changes the function of female individuals into the function of males (protopginus) or vice versa. This is closely related to the work of hormones in addition of influenced by the environment.

The investigation into the gonads of *S. macrostetholepis* Blkr. can be used as targets to determine the actual sex and the succession process ovaries into testes or vice versa. In addition, this research can be the additional information on the biological reproduction of gobies (*S. macrostetholepis* Blkr.) in the way of preservation and development of fish farming. Since, Gobi fish not only has high economic value, but also has a good taste to be consumed (Sulistino, 2012); (Willis, 2012).

**CONCLUSIONS**

From the research has been done on the relationship of the body length and the papilla with sex ratios at gobies (*S. macrostetholepis* Blkr.). It lives in the waters of Sungai Batangkuranji, Padang City, it was concluded that, The length of the body does not affect the ratio of the papilla, both females (adult females and female intersex) and male fish. The body length gobies (*S. macrostetholepis* Blkr.). Females has an average $59.18 \pm 9.06$ mm, and the average ratio of the papilla is $1.83 \pm 0.36$, while the average length of the body the male fish is $56.93 \pm 9.01$ mm, with an average ratio of $0.81 \pm 0.15$ papilla. In female fish, it is found in different stages of development, namely adult females and females intersex (the average length of an adult female body are $9.21 \pm 59.83$ mm, while the average length of the female body intersex is $58.06 \pm 8.79$ mm. The average adult female papilla ratio is $1.85 \pm 0.37$ and the average female intersex is $1.79 \pm 0.34$.

**REFERENCES**


