Growth patterns, sex ratio and size structure of nurseryfish (*Kurtus gulliveri* Castelnau, 1878) according to the lunar phase in Maro River, Merauke

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Abstract. This study was aimed to analyze growth patterns, sex ratios, gonadal maturity levels, size structures and factor conditions of nurseryfish (*Kurtus gulliveri*) which were caught at different lunar phases in the Maro River, Merauke. Fish samples were collected every month, using a gillnet, from April to September 2018. During the study, 72 individuals, consisting of 31 female and 41 male were collected. The sex ratio of fish in the lunar phase is the same, namely 1:1. Female gonads dominate maturity levels I and IV at each lunar phase, while males dominate the maturity levels of gonads II, III and V in the new moon phase. Male and female at different lunar phases generally have negative allometric growth, except female nursery fish caught in the full-phase, which have a positive allometric growth pattern. Full-weight equation of male and female *K. gulliveri* in the full-phase is log W=-4,613+2,7821 log L and log W=-5,9099+3,315 log L, while the length-weight equation for male and female *K. gulliveri* in the new moon phase is log W=-5.2155+3.0297 log L and log W=-5.0946+2.9793 log L. The range of condition factor (K) values from *K. gulliveri* in the full moon phase was 1.0135±0.15 (male), 1,0031±0.08 (female) and in the newmoon phase 1.1749±0.81 (male), 1,0044±0.098 (female), which shows the fish in good condition.

Key Words: gonadal maturity levels, condition factor, growth pattern, moon phase.

Introduction. Nursery fish, *Kurtus gulliveri* is an endemic fish species distributed only in limited areas in the southern part of Papua (including the PNG area) and the northern part of Australia. It spreads from freshwater to estuarine waters (Berra 2007a). *K. gulliveri* is a species of fish that is unique and famous for its parenting technique. Male *K. gulliveri* maintain and carry the mass of fertilized eggs by attaching to the supra-occipital that is shaped like a horn, located on the head, which is known as 'forehead brooding' (Balon 1975; Berra & Neira 2003b; Berra 2003a; Berra et al 2007b). Studies related to *K. gulliveri* have been widely carried out to elaborate some aspects regarding this species, such as: the anatomy of the digestive tract and its food (Berra & Weed 2001), the anatomy and histology of male supra-occipital hooks (Berra & Humphrey 2002), early life (Berra & Neira 2003), distribution comparison between the egg mass of *K. gulliveri* and of *Kurtus indicus,* a species living in South Asia (Berra 2003), reproductive anatomy and the relationships between: gonads and spawning season of *K. gulliveri* (Berra et al 2007b), larvae and its spawning season (Berra et al 2016), and salinity and *K. gulliveri* spawning (Berra & Weed 2017). However, less research has been conducted on the length-weight relationship of *K. gulliveri*, related to the moon phase, while the study of the relationship of length-weight is an important prerequisite and parameter in estimating the structure of length and age for managing the dynamics of fish populations, in the fishery sector (Biswas et al 2011; Paswan et al 2011; Isa et al 2012).

This research was conducted to examine the biological aspects including sex ratio, gonad maturity level, size distribution and growth pattern of *K. gulliveri* in Maro River waters, based on moon phase period, namely full moon and newmoon. Several studies related to moon phase and aquatic organisms have been carried out such as the influence of the moon cycle on the distribution and feeding activity (Quinn & Kojis 1981; Picapedra et al 2018), behavior, reproduction and spawning (Naylor 2001; Saavedra & Ferreira 2006; Horky et al 2006; Numata & Helm 2014; Takemura et al 2015), the effect of the moon phase on fish movements and catches (Libini & Khan 2012; Das et al 2015; Milardi et al 2018). However, less information was available on the fish growth patterns related to the moon phase. The results of this study are expected to add and enrich existing information so that it can be used in the management of *K. gulliveri* that live endemic in the southern waters of Papua.

Material and Method

Time and location of the study. This study was conducted from April to September 2018 in Maro River, Merauke Regency, Papua Province, Indonesia. The study site was selected based on observation on the condition of the study location. There were four stations: Station I was located at the mouth of the Maro River; Station II was located around the main port; Station III in areas without settlements and Station IV around the Matandi Harbor.

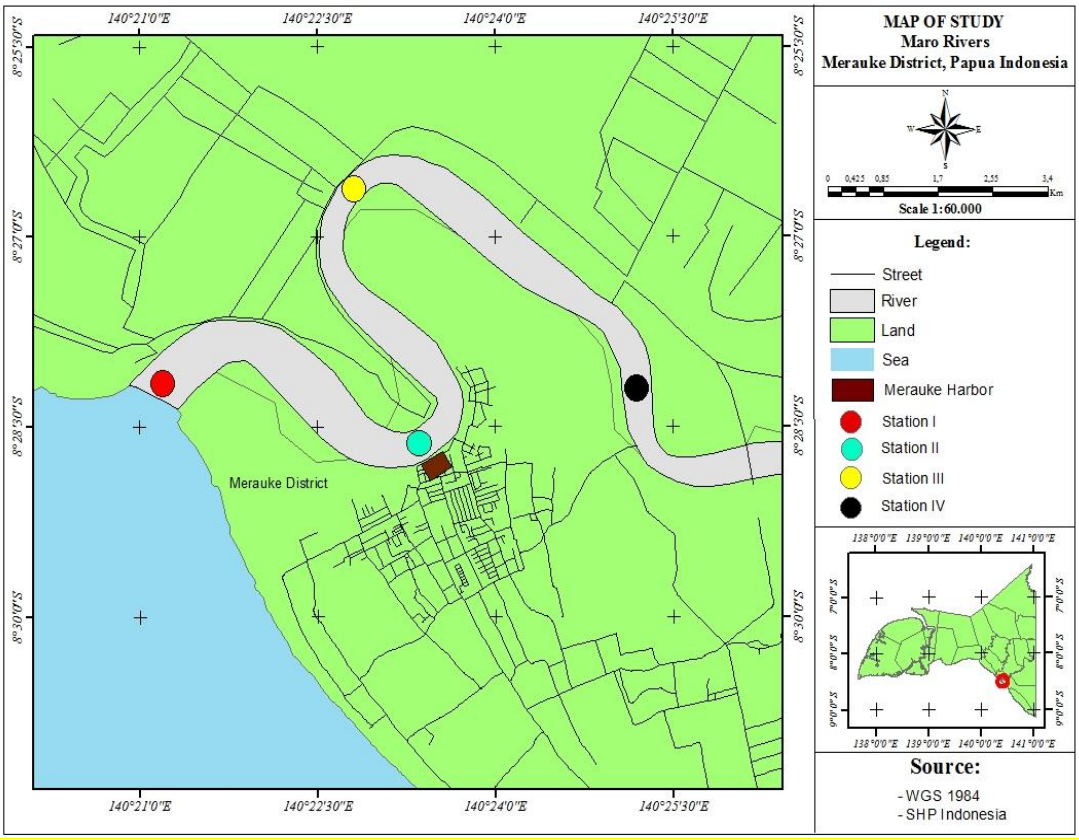

Figure 1. Location of fish sampling in the Maro River, Merauke Regency, Papua Province, Indonesia.

Fish sample handling techniques. Fish samples were collected by catching using an experimental gill drift net at each research station with a mesh size of 3, 5 and 7 inches. The length of the river from station I to station IV was 13.41 km, the width of the river was 0.87 km with the total area of 8.93 km^2 sampling station. The sample that has been obtained was then measured the total length of the individual (mm) to the accuracy of 1 mm for body weight and weight (g) to the accuracy of 0.01 g. The determination of sexuality and gonadal maturity level of *K. gulliveri* was made using modifications to Cessie (1956) and Effendie (1979) as shown in Table 1.

Table 1
Gonadal maturity level of *Kurtus gulliveri* based on morphological criteria as used by Cassie (1956) and Effendie (1979)

Maturity level	*Female*	*Male*
I	The ovary is like a long thread, at the front of the body cavity. Clear color. Sleek surface.	Testes are like shorter (limited) threads, with visible edges in the body cavity. Clear color.
II	Larger ovary size. Yellowish darker coloring. Eggs are not clearly visible to the eye.	Larger testes size. White coloring like milk. The shape is clearer at level I.
III	Ovaries are yellow, morphologically the eggs begin to appear with the eyes.	The surface of the testes appears jagged. The more white the color, the greater the testes. In a preserved state, easily broken.
IV	The ovaries are getting bigger, the eggs are yellow, easy to separate. Oil grain is not visible, filling ½ - $^2/_3$ of the abdominal cavity; urged intestine.	As at level III it seems clearer. Testes increasingly solid.
V	Ovary wrinkled, thick walls, leftover eggs near release.	The testes on the back are deflated and near the release, but still filled.

Data analysis. The relationship of fish weight and length was analyzed using the formula proposed by Hile (1963) and Le Cren (1951):

$$W = a\ L^{b}$$

Where: W=fish weight (g), L=total fish length (mm), a and b=constant.

Then it was transformed into a logarithmic form, thus forming a straight line equation as follows:

$$\text{Log } W = \log a + b \log L$$

After transforming into the logarithmic form of the original data, the values of b can be determined by using the least squares method (Akyol et al 2007) and the value of a can be retrieved through the anti-logarithm determination.

If b=3, the growth of fish shows isometric growth patterns, meaning that the increase in body length and weight is balanced. If the value of b<3 indicates a negative allometric growth pattern (minor allometric), body length increase is faster than body weight gain. Conversely, if b>3 shows a positive allometric growth pattern (major allometric), body weight gain is faster than body length increase.

To test the regression coefficient whether b=3 or not, a t-test data analysis was performed. The value of b obtained in the linear regression is significantly different from the isometric value (b=3) or allometric range (negative allometrics: b<3 or positive allometrics: b>3) when t-test (H0:b=3) with confidence level of 95% (Lawson et al 2013), expressed by the following equation:

$$t_s = \frac{(b-3)}{sb}$$

$$sb = \frac{\sqrt{[(sW/sL) - b^2]}}{(n-2)}$$

Where: t_s=student's t test, b=slope, sb=standard error of the slope, sW=variance of body weight, sL= variance of total length, n=sample size.

The strength of relationship between fish weight and length was measured using the following correlation analysis:

$$r = \frac{N(\sum \log L * \log W) - (\sum \log L)(\sum \log W)}{\sqrt{\{N(\sum \log^2 L) - (\sum \log L)^2\}\{N(\sum \log^2 W) - (\sum \log W)^2\}}}$$

Where: r = correlation coefficient, N= total number of individual species, L=total length of fish (mm), W=total weight of fish (g).

The value of r is between -1 and +1 (-1≤r≥+1). If the value of r=+1, then it is said that there is a perfect direct linear relationship between length and weight. If the value r=-1, it means that there is a perfect linear relationship which has an inverse correlation between body length and body weight. Conversely, if the value of r=0 indicates there is no linear relationship between length and weight. The strength of correlation relationship is shown in Table 2.

Table 2

Interpretation of correlation relationship (r)

Value of correlation coefficient (- or +)	*Meaning*
0.00 - 0.19	Correlation is very weak
0.20 - 0.39	Weak correlation
0.40 - 0.69	Medium correlation
0.70 - 0.89	Strong correlation
0.90 - 1.00	Correlation is very strong

The condition factor analysis is stated by Effendie (1979) using the formula:

a. Condition factor (Kn) with isometric growth pattern (b=3):

$$K_n = \frac{10^5 W}{L^3}$$

b. Relative condition factors (Kn) with allometric growth patterns (b≠3):

$$K_n = \frac{W}{aL^b}$$

Where: K = condition factor, W = weight (g), L = length (mm) and a, b = constant.

Results and Discussion

Sex ratio and gonad maturity level. During the sampling, carried out from April to October 2018, 72 *K. gulliveri* individuals were collected, consisting of 41 male and 31 female individuals (Table 3). The highest catches of *K. gulliveri* in the newmoon phase were of 45 individuals, whereas in the fullmoon phase there were of 27 individuals. Male fish are generally caught more than females during the full and newmoon phases. The ratio of male and female fish based on the chi-square test shows that the full moon phase X^2 is 0.07 and the X^2 value in the new moon phase is 1.81.

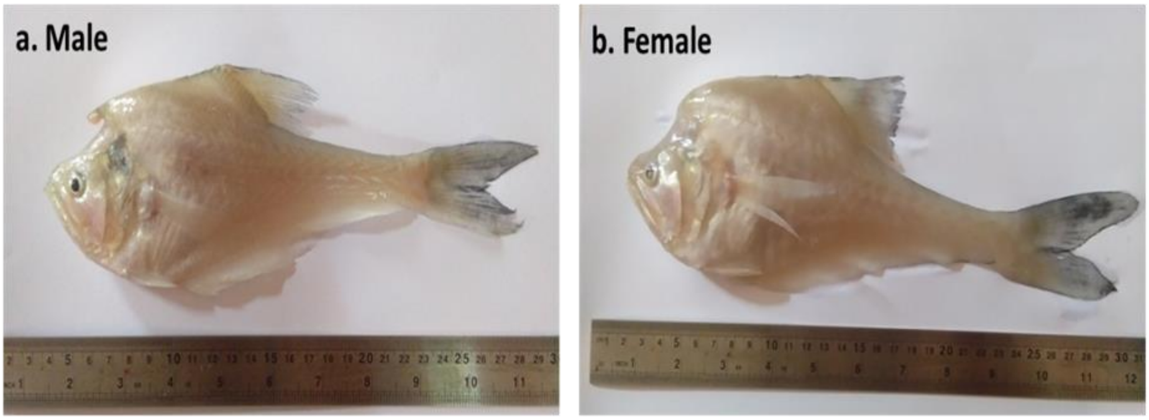

Figure 2. *Kurtus guliveri* (a. Male; b. Female).

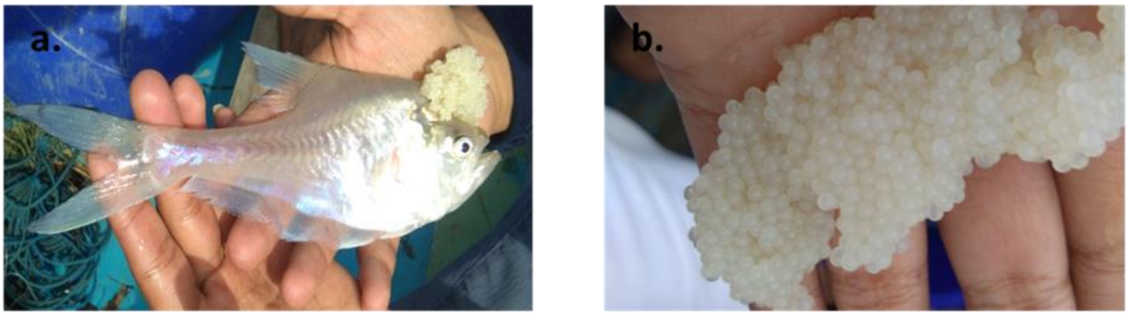

Figure 3. a. Male *Kurtus gulliveri* take care of eggs on supra-occipital hooks (forehead brooding); b. The mass of *Kurtus gulliveri* eggs.

Table 3

The sex ratio of *Kurtus gulliveri* during different lunar phases

Lunar phase	*Male*	*Female*	*Male: female ratio*	*X^2 test and X^2 table (α=0.05; db=1)*	*Total (Ind)*
Fullmoon	14	13	1.00:0.93	0.07<3.84	27
Newmoon	27	18	1.00:0.67	1.82<3.84	45
Total	41	31	1.00:0.76	1.40<3.84	72

Df-degree of freedom, Ind-individual.

The gonad growth rate of *K. gulliveri* during the different phases of the study period, presented in Table 4, shows that, overall, female fish dominated at GML I (58%), GML II (16.1%), GML III (6.5%) and GML IV (19.4%), in contrast to male fish in GML I (75.6%), GML II (12.2%) and GML III (12.2%), no GML IV was found during the study period.

Table 4

Distribution of *Kurtus gulliveri* gonad maturity level during the study period

GML	*Fullmoon*		*Newmoon*		*Total (%)*		*Total (ind)*
	Male	*Female*	*Male*	*Female*	*Male*	*Female*	
I	13	8	18	10	31 (75.6)	18 (58)	49
II	-	2	5	3	5 (12.2)	5 (16.1)	10
III	1	1	4	1	5 (12.2)	2 (6.5)	7
IV	-	2	-	4	0 (0.0)	6 (19.4)	6
Total	14	13	27	18	41	31	72

GML-gonad maturity level.

Size structure. Distribution of *K. gulliveri* size during different moon phases informed that length of male fish during the fullmoon phase ranged from 53-395 mm with a body weight ranging from 1.8-470 g (average: 98.2 g). In the newmoon phase, the total length of male fish ranged from 40-265 mm with body weights ranging from 16 to 143 g (average: 52.04 g). Conversely, the total length of a female fish in the fullmoon phase ranges from 150-365 mm with a body weight ranging from 19-352 g. The total length of females caught in the newmoon phase ranges from 130-380 mm with a body weight ranging from 15-423 g (Table 5). Based on the moon phase, the average value of both the total length and body weight of *K. gulliveri* is higher in the fullmoon phase compared to the newmoon, whereas in terms of *K. gulliveri* sex, the average total length and body weight of this species is higher in male fish than in female fish (Figure 4).

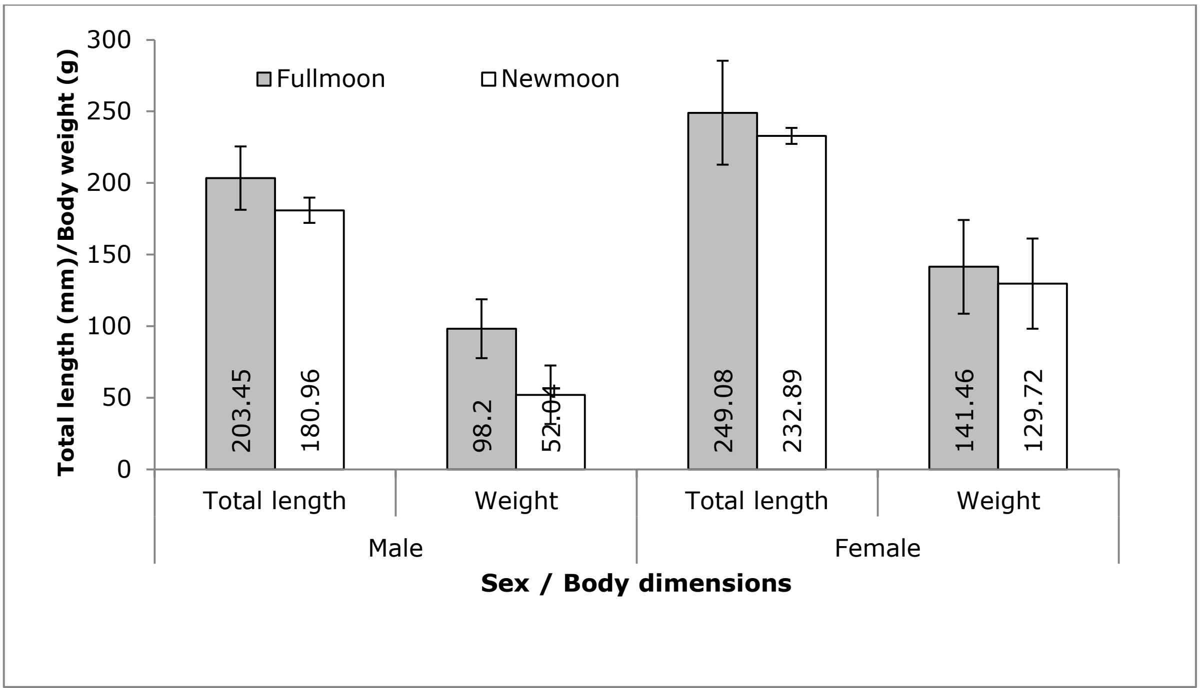

Figure 4. The average value of total length and body weight of *Kurtus gulliveri*, caught at fullmoon and newmoon phases, in the estuary of the Maro River, Merauke, separated by sexes.

Based on the length and weight class intervals for male and female fish, the frequency of individual male fish was plotted in the two moon phases. In the fullmoon phase, the length classes were 181-292 mm with a frequency of 6. In the newmoon phase, the length classes 192-229 mm were obtained, with an individual frequency of 10. The highest individual frequency in the body weight class of 1.8-95.7 g was 12 in the newmoon phase. Conversely, in the newmoon phase, a frequency of 7 was obtained at class intervals of 16-37 g (Figures 5 and 6).

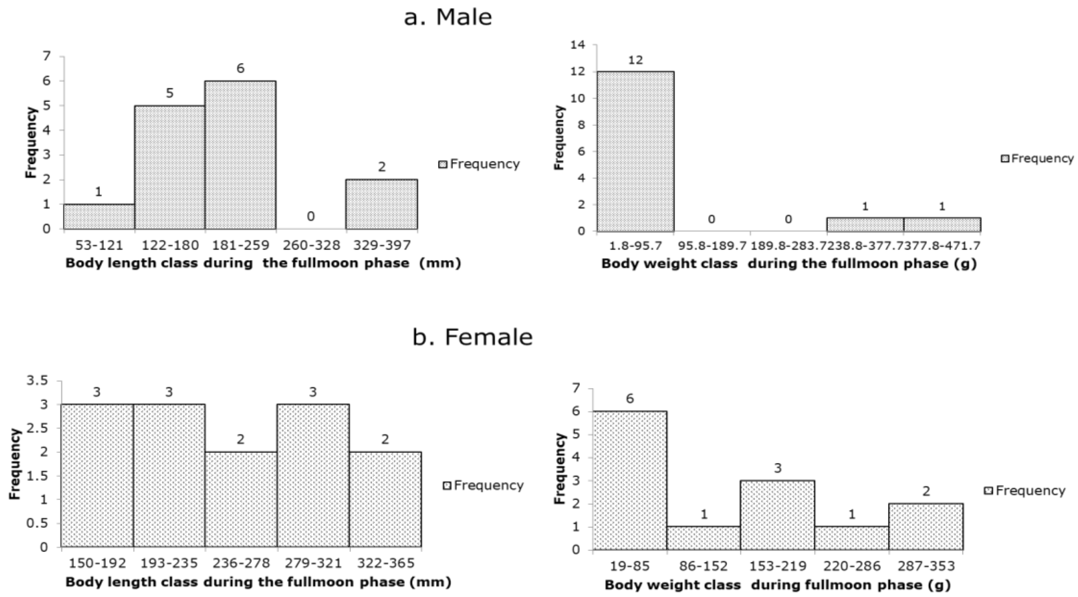

Figure 5. Class of frequency distribution of total length and body weight of *Kurtus gulliveri* caught in the fullmoon phase.

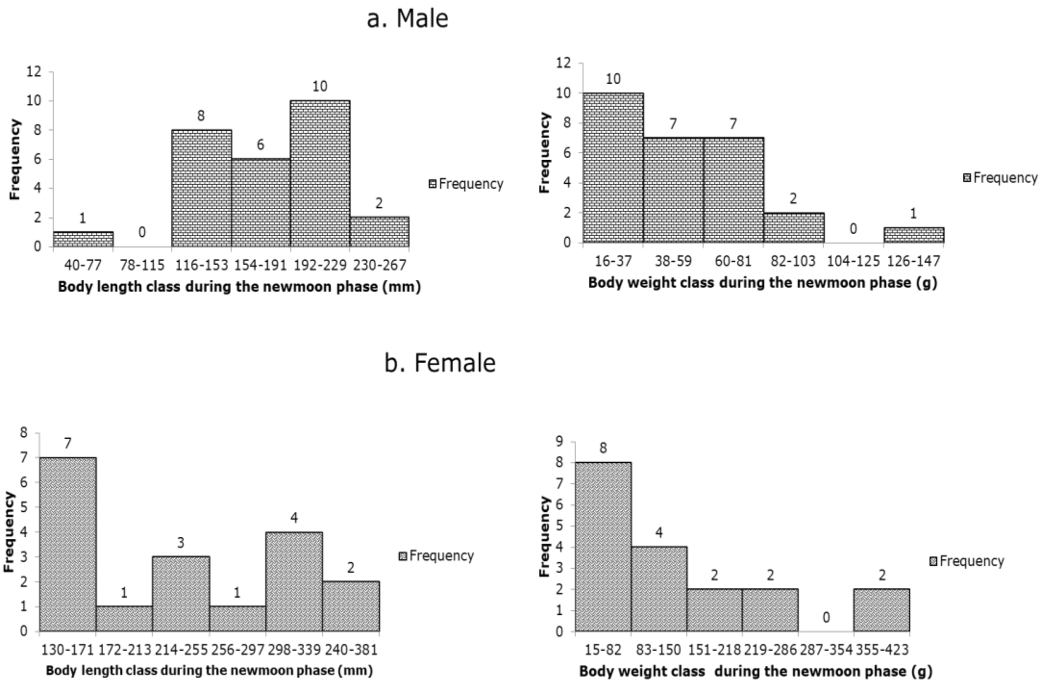

Figure 6. Class frequency distribution of total length and body weight of *Kurtus gulliveri* caught in the newmoon phase.

Length-weight relationship. The results of the analysis of the relationship between length and weight of *K. gulliveri* in different moon phases used in determining the growth patterns showed that the analyzed *K. gulliveri* were of total lengths ranging from 53-395 mm and their body weight range between 1.8-470 g for males during the period of the research and for females with a range of total length between 130-380 mm and body weight range between 15-423 g (Table 5). The length-weight relationship of male *K. gulliveri* weight in the moon phases follows the equations $W=0.0000243\ L^{2.782}$ for the fullmoon phase and W=0.0000061 $L^{3.0297}$ for the newmoon phase. Conversely, the equation for female *K. gulliveri* growth patterns in the fullmoon phase was W=0.0000012 L^{3315} and in the newmoon phase it was W=0.000008 $\mathbf{L}^{2.979}$ (Figure 7 and Figure 8). The results of the t-test analysis of the b value of male and female *K. gulliveri* in different moon phases with 95% confidence interval (α=0.05) showed a value of b<3, indicating a negative allometric patterns found in males during the fullmoon phase and in female *K. gulliveri* during the newmoon phase. Conversely, a positive allometric growth pattern (b>3) was found in male fish during the newmoon phase and in female fish during the fullmoon phase.

A graph with a linear pattern in male and female fish shows that larger males gained a greater body weight (Figure 7 and Figure 8). This can be interpreted even though the body size is the same length, but the male is bigger than the female based on body weight. The strength of the relationship between the length and weight of *K. gulliveri*, based on the correlation value r, indicated a very strong correlation of male fish in the fullmoon phase (r=0.9929) with female fish in the fullmoon phase (r=0.9967) and in the newmoon phase (r=0.9962). In contrast, male fish in the fullmoon phase had moderate correlation strength (r=0.4214) with male *K. gulliveri* in the newmoon phase (Table 5).

Table 5

Results of analysis of the length-weight relationship of *Kurtus gulliveri* at different lunar phases

Sex	*Parameter*	*Lunar phase*	
		Fullmoon	*Newmoon*
Male	N	14	27
	TL range (mm)	53-395	130-265
	Average TL±SE	203.45±22.17	187.04±6.94
	W range (g)	1.8-470	16-143
	Average W±SE	98.2±36.28	52.04±5.67
	a	0.0000243	0.0000061
	b	2.7821	3.0297
	r^2	0.9858	0.9419
	r	0.9929	0.9706
	Relationship strength	correlation is very strong	correlation is very strong
	t-test	0.404	-0.25
	t-tab (0.05, n)	2.145	2.05
	Growth pattern	negative allometrics	positive allometric
	W = aL ^{b}	0.0000243L ^{2782}	0.0000061L $^{3.0297}$
Female	N	13	18
	TL range (mm)	150-365	130-380
	Average TL±SE	249.08±20.53	232.89±20.46
	W range (g)	19-352	15-423
	Average W±SE	141.46±32.78	129.72±31.54
	a	0.0000012	0.000008
	b	3.315	2.9793
	r^2	0.9934	0.9923
	r	0.9967	0.9962
	Relationship strength	correlation is very strong	correlation is very strong
	t-test	-0.776	0.046
	t-tab (0.05, n)	2.16	2.10
	Growth pattern	positive allometric	negative allometrics
	W = aL ^{b}	0.0000012L $^{3.315}$	0.000008L $^{2.979}$

TL-total body length; W-body weight; SE-standard error; N-number of samples.

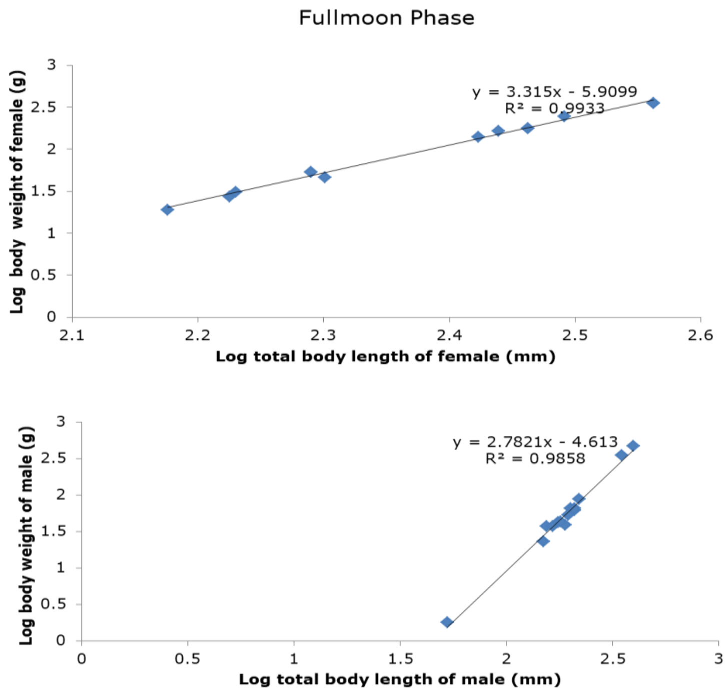

Figure 7. Length-weight relationship of *Kurtus gulliveri* caught during the fullmoon phase.

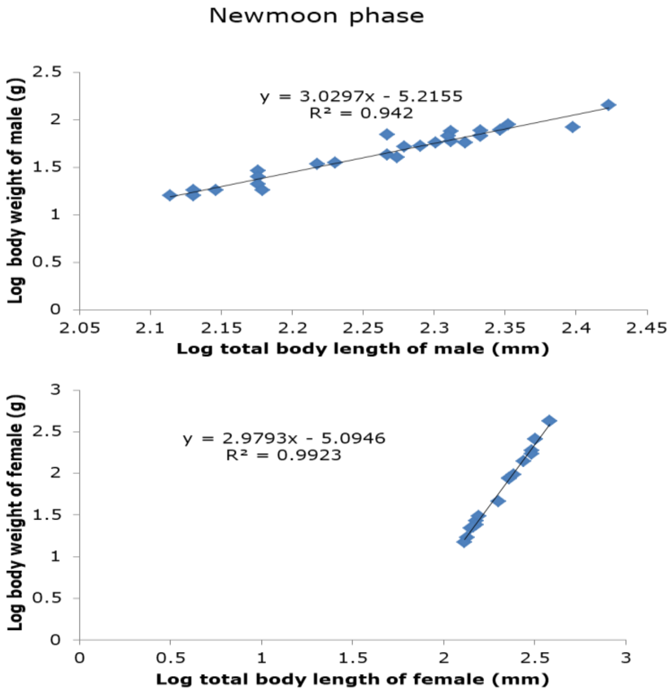

Figure 8. Length-weight relationship of *Kurtus gulliveri* caught during the newmoon phase.

Condition factor. The highest condition factor, ranging from 0.4359 to 4.4680, was observed for male fish in the newmoon phase and the lowest, ranging from 0.8804 to 1.1032, was observed for female *K. gulliveri* sh in the fullmoon phase (Table 6).

Table 6

Condition factors of *Kurtus gulliveri* at different lunar phases

Description	*Condition factor (K)*			
	Fullmoon		*Newmoon*	
	Male	*Female*	*Male*	*Female*
K range	0.7341-1.2271	0.8804-1.1032	0.4359-4.4680	0.7979-1.1291
Average of K±SD	1.0135±0.15	1.0031±0.08	1.1749±0.81	1.0044±0.098

SD-standard of deviation.

Discussion. The X^2 value of male and female *K. gulliveri* was obtained from different moon phases, the two values were compared with chi-square table with ɑ of 0.05 and db=1 got a value of 3.84 as shown in Table 3, indicating the calculated X^2 value $<X^2$ value from thee chi-squared table for different moon phases. The sex ratio of males and females was balanced. This is similar to Sulistiono et al (2009), which stated that the sex ratio of male and female in a fish population is 1:1. This is an ideal condition, although based on the catch data, more male fish are caught than female fish in different moon phases. This may be related to the separation of some forms of adulthood through habitat preferences and migration or differences in behavior between sexes, making one sex more susceptible to be captured than the other (Jega et al 2017). Ongkers et al (2016) explained that the same sex ratio is ideal for reproduction allowing one male fish to fertilize one female fish. This is in agreement with the uniqueness of the male *K. gulliveri* species that are unique in terms of the nurturing behavior of caring for and maintaining the fertilized eggs in the head in the form of a supra-occipital hook on the head (He et al 2016), in addition, male fish are more susceptible to be caught against gill nets due to these supra-occipital hooks. Berra et al (2007b) further explained that unclear sexual differences related to female spawning behavior, habitat preferences, social interactions, seasonal movements, may affect the sex ratio of *K. gulliveri*.

Based on the research time from March to September, gonad maturity levels at level I are more commonly found in males and females in different moon phase conditions and it is also found that gonad maturity levels at levels III and IV which indicate that spawning has occurred. Gustiarisanie et al (2017) described that the ongoing spawning season is characterized by the presence of fish that have mature ripples, while the spawning grounds in waters are determined by the discovery of male fish and mature female fish together. Berra et al (2007b) described that *K. gulliveri* species have a long reproductive season of the month from June to November, so there can be found variations in the level of maturity of the gonads. Differences in the level of gonad maturation found in different moon phases indicate that the moon phase exerts influence on the level of endocrine control of gonad maturation that stimulate events which will produce ovulation in the next phase of the moon (Ali 1992).

The size structure of the *K. gulliveri* found at different lunar phases informed that the length size of both males and females in average is longer than the *K. gulliveri* caught at newmoon phase. Similar trend is also observed in terms of the body weight. Hart & Reynolds (2002) explained that body length almost always increases with time, whereas weights may go up or down during certain time intervals depending on the influence of various factors that affect deposition and mobilization of body material. The influence of the moon on tidal changes determines fluctuations of regional water temperatures, turbidity, salinity (especially in estuarine areas) and also influences the availability of food (Ali 1992), this also influences the growth of fish i.e. length and weight. The length of *K. gulliveri* during the study for male fish ranged from 40-395 mm and the weight ranged between 1.8-470 g, while the standard length of females ranges from 130-380 mm and body weight ranges from 15-423 g. The length of *K. gulliveri* has been reported by several studies such as Bera & Neira (2003), who found males carrying eggs at their head with a length of 190 mm in the Ajkwa estuary region, Timika (Papua Province, Indonesia), in Berra et al (2007b), the length range for the Adelai River (Australia) males ranged from 175-240 mm and for the females it ranged from 204-304 g, while the highest value was reported to be 590 mm (Weber 1913).

An analysis of the relationship between length and weight was conducted to determine the growth pattern of fish species. It is assumed that growth patterns may be determined by comparing the value of b from the t-test analysis (Syahrir 2013). *K. gulliveri* shows that in general the growth pattern of *K. gulliveri* in different moon phases has a negative allometric growth pattern (b<3) which means that length growth is faster than weight growth, so that fish become slimmer because they become longer (Riedel et al 2007). However, female *K. gulliveri* in the fullmoon phase have positive allometric growth patterns (b>3) showing a weight growth faster than the length growth (Pauly 1984; Froese 2006). Chukwu & Deekae (2010), stressing that the fish weight, associated with metabolism in all species, depends on the environment in which they live. The difference in the value of "b" in the same habitat may be related to many factors (Okomoda et al 2018) fish growth being influenced by both internal and external factors. Internal factors are generally difficult to be controlled. They include heredity, sex, age, parasitic population, and diseases, while external factors in fish are influenced by factors such as: the temperature, diet, season, habitat, intensity of light entering the water column in fish foraging activities and competition in the food chain (Paswan et al 2011; Rahardjo et al 2011; Wujdi et al 2012; Jusmaldi & Hariani 2018; Omotayo et al 2018). Correlation coefficient "r" shows the strength of the relationship between length and weight of *K. gulliveri* during the study period, suggesting that increasing fish weight will affect its increase in length (Jusmaldi & Hariani 2018), except for male *K. gulliveri* caught in the newmoon phase, having a moderate correlation (r=0.4214). The moon cycle is related to the tidal cycle, which has a strong relationship with fish foraging behavior (Ono & Addison 2009). Takemura et al (2015) described the lunar cycles involving a change periodically repeated at intervals of one month, affecting the biological activity peaking around the phases of the moon, which can be attributed to the fish in search for food or reproduction opportunities.

The "K" condition factor is another important parameter derived from length and weight relationship data, suggesting fish growth patterns, including obesity, and is also useful in evaluating the importance of various spawning areas (Ribeiro et al 2004; Rahardjo & Simanjuntak 2008; Syahrir 2013). In summary, the condition factor is an efficient instrument showing changes in fish conditions throughout the year (Rahardjo & Simanjuntak 2008). This parameter is a measure of how well the fish are in their environment, allowing for the mutual conversion of fish length and weight (Chukwu & Deekae 2010). Furthermore, Effendie (2002) explained that the condition factor is a value that may be used to indicate the state of fish both in terms of physical capacity for survival and reproduction. Based on the range of condition factors shown in Table 6, all *K. gulliveri* caught have a variable body curvature, based on a range of "K" values between 1-3 (Effendie 2002). The value of the condition factor of *K. gulliveri* male or female caught in different phases of the moon indicate that the fish are in good condition and also that the habitat conditions are favorable to the fish growth (Sentosa & Satria 2014).

Conclusions. *K. gulliveri* caught during the study period had a ratio of 1:1, female fish dominated GML I and GML IV at two different phases of the moon. Male fish in the newmoon phase dominated GML II, GML III and GML V, whereas in the fullmoon phase only GML I and GML III were found. Generally the growth of *K. gulliveri* in different lunar phases had negative allometric patterns, except for females in the fullmoon phase, which had positive allometric growth patterns. The equation of the length-weight relationship of male *K. gulliveri* in the fullmoon phase was: log W=-4.613+2.7821 log L and for female: log W=-5.9099+3.315 log L, whereas the equation of length-weight in the newmoon phase for male *K. gulliveri was*: log W=-5.2155+3.0297 log L and for female: log W=-5. 0946+2.9793 log L. *K. gulliveri* grew well in the Maro River, during the different phases of the moon, with the value of the condition factor close to 1.

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