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Reproductive parameters of Reticulate Knife fish, *Papyrocranus afer* (Gunther, 1868) from Lekki Lagoon, Lagos Nigeria

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Sizes at maturity (L_{m50}), fecundity, sex ratio and gonado somatic indices for *Papyrocranus afer*, collected from Lekki Lagoon, were studied from January 2010 to April 2012. In total 1154 specimens with standard lengths, 5.2-75.9 cm (34.86±17.2cm) and body weight, 7.9-1, 958.8g (249.12 ± 28.56g) were collected by means of artisanal passive and active fishing gears (traps, long lines and nets. Sexes of fish specimen were determined macroscopically after dissection and the length at which 50% of the fish population reaches sexual maturity (L_{m50}) was considered as length at sexual maturity. Fecundity was determined by total counts of eggs; sex ratio by proportion of male to females, and gonadosomatic index (GSI) was expressed as gonad weight as percentage of total body weight. Results indicated that the most frequently caught fish size was 34.5cm; while sizes at maturity were 49.1cm (males) and 53.4cm (females).There was significant difference in estimated sex ratio X^2 (1) = 32.21, p < 0.0001, 1:0.6 (Male: Female). Fecundity was low (mean 49 ± 17 eggs for a fish 52.86 ± 7.53 cm); it increased with fish size (r = 0.71). Higher GSI during rainy season with peak in July (female: 0.44 ± 0.14; male 0.22 ± 0.01 %) indicated seasonal spawning. Low fecundity and annual spawning underlined the need for sustainable exploitation and management of this species in Lekki Lagoon.

Keywords: *Papyrocranus afer*, Gonad maturity, Fecundity, Sex ratio, Breeding season Lekki Lagoon.

INTRODUCTION

The knifefishes belong to Family Notopteridae, which comprises four genera namely *Papyrocranus, Xenomystus, Chitala* and *Notopterus;* represented by ten species (Robert, 1992). Two genera, *Papyrocranus* and *Xenomystus* are endemic to West and Central Africa while the remaining two (*Chitala* and *Notopterus*) are endemic to India and South Asia (Robert, 1992). All species are found living in freshwater environments (Robert, 1992; Inoue *et al.,* 2009, Hilton, 2002). The Notopteridae is represented by two genera, (*Papyrocranus* and *Xenomystus)*, each with a single species, *P. afer* and *X. nigri* in Nigeria.

The African reticulate knifefish, *Papyrocranus afer* (Gunther, 1868) has no commercial importance in Nigeria as food fish. Its true potential lies in the use of its juveniles as ornamental fish. However, the supply of this species in Nigerian Ornamental Fisheries relies upon fish collection from the wild with its associated problem of inadequacy, seasonal availability, habitat degradation, loss of catchment area, overfishing, heavy post-harvest lost, pollution (Olaosebikan, 2005) and possible introduction of pathogens/ diseases into culture enclosures. In the prevailing trends of growing local and international ornamental fish demands, the sustainability of the fish stocks is not guaranteed. There is paucity of documented data on reproductive biology of *P. afer* in Nigeria although such information represents a critical element of the biological basis for management of this species. Alves and Minte-Vera (2012) opined that scarcities of reproductive biology data have lead to stock overexploitation and management failure in many countries. Knowledge about species reproductive biology is essential to promote its exploitation in a sustainable way, avoiding depletion of stocks (Rossoni *et al.*, 2010).

Fish reproductive biology plays an important role for fishery management, especially in developing countries. Such information constitutes essential component of the management of any fish stock (Morgan, 2008; Trindade-Santos and Freire, 2015). Factors used in estimating the reproductive potential of fish stocks included sex ratio, size at maturity, fecundity and gonadosomatic index (Jennings *et al.,* 2001; Morgan, 2008; Lowerre-Barbieri *et al.,* 2015). Size at sexual maturity is a particularly important parameter used to assess and evaluate the impact of fishing mortality on spawning stock biomass, to determine levels of optimum fishery yield and estimate reproductive load of fish species (Polovina, 1987). Sex ratio, fecundity and gonadosomatic index are important information to assess the structure of populations, their reproductive potential as well as outset and period of spawning. Such information is surprising not available for most ornamental fish species in Nigeria. The fragmented information available (NIFFR, 2002; Mbawuike and Pepple, 2003; Mbawuike, and Ajado, 2005; Ukaonu *et al.,* 2011) does not include information on aspects of their reproductive biology. Hence, this study aimed to examine size at maturity, fecundity, gonadosomatic indices as well as sex ratio of *Papyrocranus afer* from Lekki Lagoon for sustainable use and exploitation of this species.

MATERIALS AND METHODS

Study Area

Lekki Lagoon lies between longitude 4˚00' and 4˚15' E and latitude 6˚25' and 6˚37' N (Figure 1). The lagoon is fed by River Oni discharging into the north-eastern part, and Rivers Oshun and Saga discharging into the north-western parts of the lagoon. It is bounded by Bight of Benin of the Atlantic Ocean in the south, and opens into the sea via the Lagos Lagoon and Lagos Harbour. It has a surface area of about 247 square kilometers and it is mostly shallow (less than 3.0 m deep) the maximum depth being 6.4 meters (Emmanuel and Osibona, 2013).

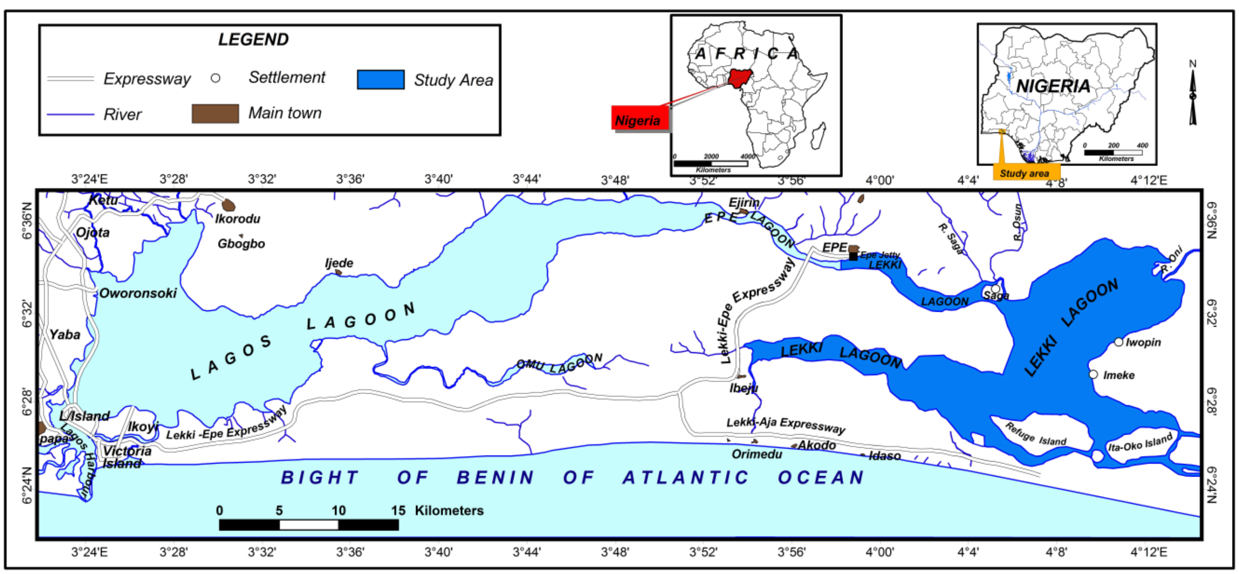

Figure 1: Map of lagoon complex of western Nigeria showing study area (■). Inserts: Map of Africa showing location of Nigeria, Administrative Map of Nigeria showing location of study area.

Collection of *Papyrocranus afer*

Samples of *Papyrocranus afer* were obtained monthly from catches of fishermen from Lekki Lagoon at Epe Jetty from January 2010 to April 2012. Fishermen exploit fish resources of Lekki Lagoon with a wide variety of artisanal gears (active and passive): such as traps (circular traps, bamboo traps, and trigger traps), long lines (baited and foul hooking), hooks number 7 - 14 as well as various nets including set nets (gill nets, drift net) throw net (cast nets), dip net, left nets and clap net (single and twin clap net). Collected specimen were transported in ice-chest containing ice in order to keep them fresh, to Fisheries Research Laboratory, Zoology

Department, University of Ibadan, Ibadan, Oyo State, Nigeria, and preserved by deep freezing (-4^{o}C) prior to examination.

Laboratory Procedures

The standard length (cm) and weight (g) were recorded for each of the specimens using fish measuring board and a digital weighing balance. Sex of the specimens was determined by examination of the gonads after dissection; gonads were removed, weighed and examined morphologically and microscopically. The size at maturity (L_{m50}) was taken as the length at which 50% of the males and females were found with matured gonads (Tsikliras *et al.,* 2013). Maturity state of the species was determined according to Agostinho *et al*. (1991). The sex ratio of male and female was calculated based on the proportions of males and females in the samples collected. Specimens collected during 2010 were not included in the analysis of sex ratio because of the initial challenges in differentiating males and female. Total Fecundity determination was done by counting total number of eggs in ripe ovaries. The linearity of fecundity–length and fecundity- weight relationship was determined using the equation reported by LeCren (1951):

Log Y = a + b Log X

Where Y = fecundity, X = length (cm) or weight of fish (g) depending on the relationship a and b = constants using ripe fish.

Correlation coefficient r was determined from regression analysis.

The monthly gonadosomatic index (GSI) was calculated using the formula reported by Jahan *et al.* (2014) as follows:

$$GSI = \frac{\text{Gonad weight}}{\text{Fish weight}} \times 100$$

The linearity of the GSI–weight relationship was determined using the equation reported by Jahan *et al.* (2014):

Log Y = a + b Log X

Where

Y = gonadosomatic index

X = weight of the fish

a and b = regression constants.

The diameters of eggs were measured (mm) under a calibrated binocular-microscope with ocular micrometer

Statistical Analysis

Data were subjected to descriptive statistics using Microsoft Excel Statistical Tool pack (2010). Pearson correlation coefficient (r) was used to test the relationship between fish length and fecundity and gonadosomatic indices and fish size. An Independent t-test was conducted to compare mean sizes of male and female. The deviation of sex ratio from the expected 1:1 was determined using Chi-square on Graph Pad Prism Version 5.00. Maturity curve were plotted using Origin O software.

RESULTS

Overall 1154 specimens of *Papyrocranus afer* with standard lengths 5.2-75.9 cm (34.86 ± 17.2cm) and body weight 7.9-1, 958.8g (249.12 ± 28.56g) were collected. Matured specimens had characteristic urinogenital papillae prominent in male but vestigial in female (Figure 2). During the study a total of 1154 specimens were examined. Sexual discrimination was only possible in 626 specimens. Matured specimens had characteristic urinogenital papillae prominent in male but vestigial in female (Figure 2). The 626 specimens comprised 384 males and 242 females Table 1. Males varied 5.3 – 66.9 cm (32.98 ± 9.54) standard lengths and weighed 10.9 – 1359 (214.83 ± 196.82). Females ranged 11.5 – 75.1 cm (46.20 ± 11.26) in standard length and weighed 21 - 1958.3 (520.91 ± 201.82) (Table 1). There was a significant difference in the mean sizes of male (39.28 ± 9.54) and female (46.20 ± 11.26); t (533) = -17.1, $p < 0.001$. The most frequently caught fish size was 34.5cm; while sizes at maturity were 49.1cm ± 5.1 cm (39.9 cm - 66.7) in males and 53.4cm ± 7.2 cm (41.2 cm – 75.9 cm) in females. Matured males are smaller than females. Maturity Ogive showed indicated the Lm_{50} for the sample for collected (Figure 3).

Estimated lengths at 50 % sexual maturity corresponding to maximum attainable lengths in both sexes were greater than 60 %. There was significant difference in estimated sex ratio X^2 (1) = 32.21, $p < 0.0001$1:0.6 (Male: Female). Fecundity ranged from 15 to 92 eggs with mean of 49 ± 6.7 eggs for a fish of 41.6 – 75.9 cm standard length (mean length = 52.86±7.53) and 344-1958.3g (mean = 875.01 ± 357.03g). Logarithmic fecundity-length and fecundity-weight relationships were represented by the following regression equations (Figures 5a and 5b)

Y=1.8895X – 1.5836 (r=0.71; p<0.05)

·Y= 1.317X – 0.1266 (r=0.75; p<0.05).

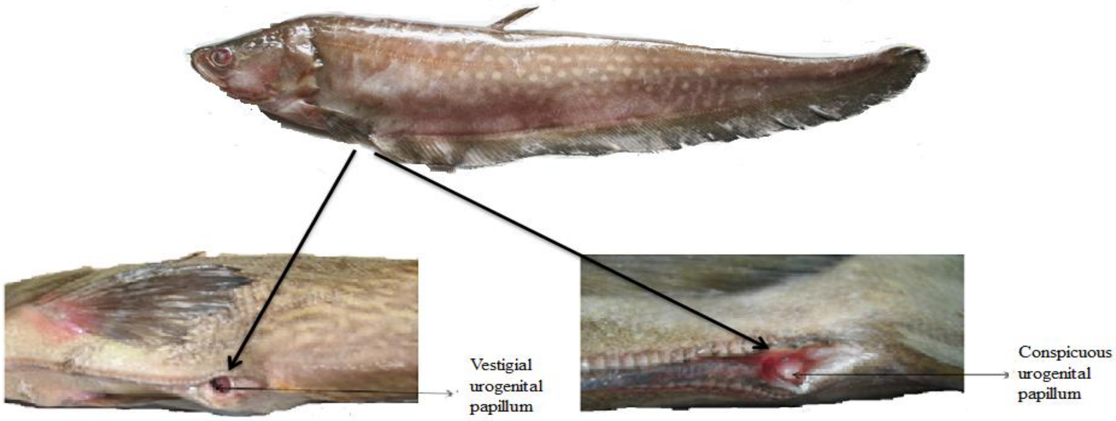

Figure 2: *Papyrocranus afer* showing vent with urogenital papillae

Table 1: Monthly variations of male and female *Papyrocranus afer*

Year 2011	**Month**	**Male (Number identified)**	**Female (Number identified)**	**Statistics**			
				Male		**Females**	
				Standard length (cm)	**Wet weight (g)**	**Standard length (cm)**	**Wet weight (g)**
	Jan	26	11	9.74 – 46.9 (30.1 ± 7.24)	23.1 - 365.6 (169.51 ± 67.68)	25.8 - 64.4 (45.97 ± 10.47	117.6 – 1003.8 (355.20 ± 213.12)
	Feb	19	13	5.3 – 42.4 (24.82 ± 11.11)	10.9 – 275 (111.92 ± 84.1)	29 – 47.5 (39.14 ± 6.36)	134.6 – 360.8 (209.31 ± 74.55)
	Mar	22	11	9.8 – 50.2 (26.6 ± 10.74)	19.1 -226 (118.82 ± 56.52)	11.7 – 65.5 (36.74 ± 11.33)	21 – 294.4 (178.32 ± 63.81)
	Apr	30	21	11.6 – 55.4 (34.1 ± 10)	21.1 – 835.6 (200.47 ± 155.64)	33.7 – 66.7 (47.36 ± 10.1)	166.8 – 1352 (583.47 ± 360.24)
	May	36	20	9.7 – 65.6 (32.52 ± 13.88)	19.1- 1359 (259.87 ± 292.33)	12.5 61 (34.7 ± 16.86)	21.1 – 1142.3 (363.68 ± 341.1
	Jun	28	17	14.6 - 61. 1 (37.5 ± 17.9)	22. 1 – 1061.8 (282.11 ± 78. 63)	36.1 – 61.3 (48.74 ± 6.12)	217.4 – 1050 (578.73 ± 211.68)
	Jul	19	15	27 – 66.9 (36.76 ± 8.08)	69.7 – 1352 (274.04 ± 196.74)	41.3 – 66.7 (50.75 ± 6.57)	216 -1312.4 (615.31 ± 283.45)
	Aug	32	29	13.8 – 61 (33.8 ± 10.3)	23.4 – 1145.7 (274.87 ± 230.98)	33.3 – 67.4 (46.89 ± 9.67)	48.6 - 1453.8 (534.85 ± 355.38)
	Sep	38	28	9.6 – 55.4 (33.09 ± 9.16)	19. 1 – 835.6 (206.16 ± 158.20)	36. – 66.6 (47.23 ± 9.21)	216.8 – 1636.7 ± 362.62)
	Oct	27	19	21.4 – 65.3 (34.27 ± 6.21)	97 – 1074.6 198.97 ±159.33)	36.8 - 69.8 (51.2 ± 11.4)	47. 4 - 1320 (520.41 ± 268.5)
	Nov	31	16	23. 8 – 60.3 (34.23 ± 3.21)	98.1 – 1042.1 (156.2 ± 91)	34.4 – 67.8 (48.23 ± 9.94)	44.6 – 1372 (569.67 ± 298.59)
	Dec	29	18	26.6 – 55.3 (33.7 ± 5.50)	68.3 – 840.4 (191.6 ± 124.12)	27. 8 – 75.1 (49.93 ± 9.79	68.3 – 1958.3 (616.84 ± 347.98)
Year 2012	Jan	20	11	10.3 – 41.7 (30.25 ± 9.18)	22 – 353 (153.64 ± 94.56))	29.8 – 61 (42.34 ± 10.76)	99.1 - 1142.3 (439.68 ± 275. 22)
	Febr	12	6	27.5 -38. 9 (34.23 ± 3.82	100.7 ± 292.3	40.7 – 66.8 (53.55 ± 10.43	352.6 – 1444.4 (794.43 ± 425.67
	Mar	9	4	32.6 – 48.8 (42.86 ± 5.87)	143.6 - 628.5 (407.82 ±169.08)	43. 9 – 52.3 (49.82± 3.9	568.2 – 835.4 (659.63 ± 119.78)
	Apr	6	3	29.3 - 50.1 (42.53 ± 9.4)	120.1 – 70.8 (432.88 ± 244.34)		49.1 – 61.1 (56.1 ± 6.27)
	Total	384	242				

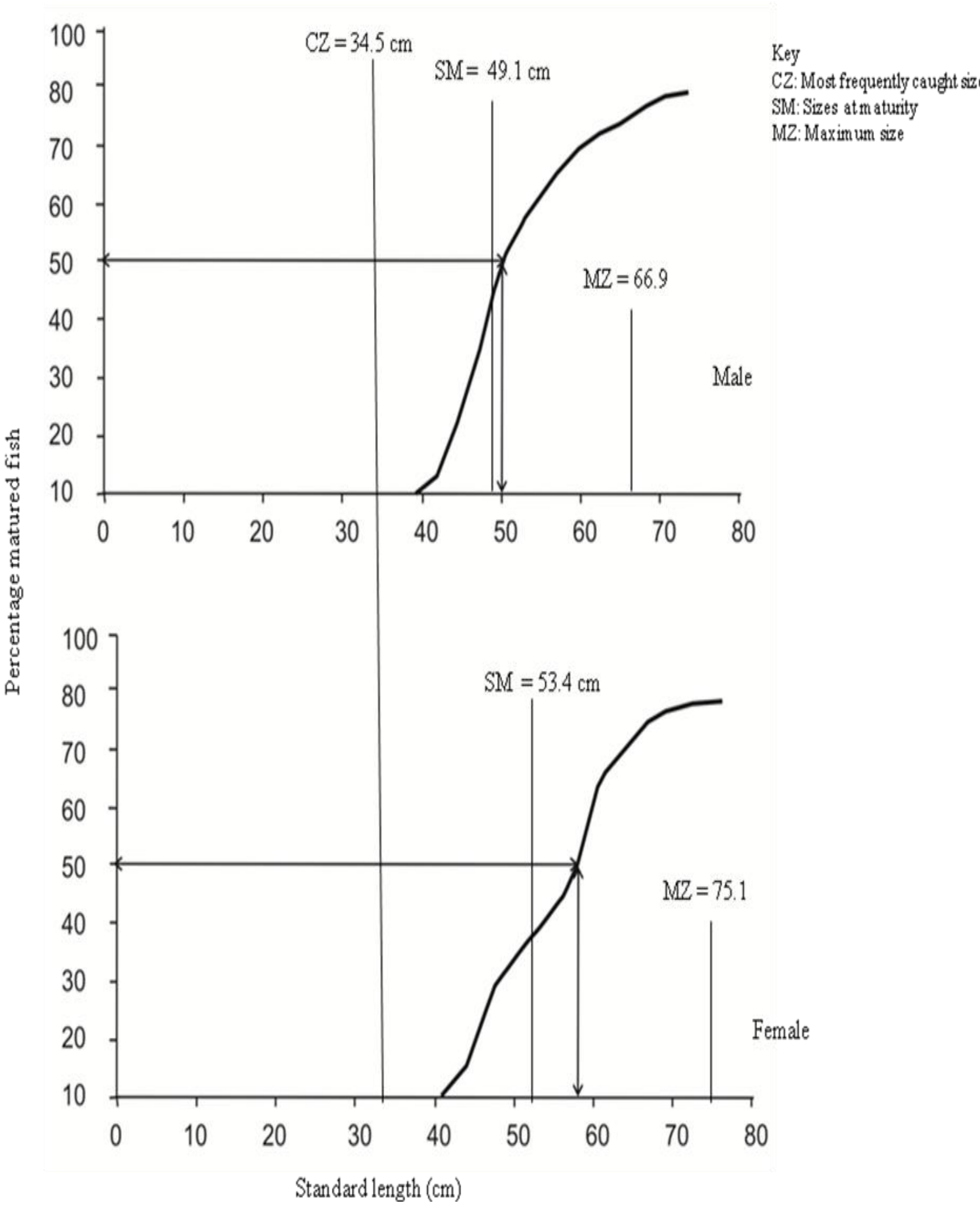

Figure 3: Maturation curve (L_{m50}) of *Papyrocranus afer* from Lekki Lagoon

The most frequently caught fish size was 34.5cm; while sizes at maturity were 49.1cm ± 5.1 cm (39.9 cm - 66.7) in males and 53.4cm ± 7.2 cm (41.2 cm – 75.9 cm) in females. Matured males are smaller than females. Maturity Ogive showed indicated the L_{m50} for the sample for collected (Figure 3).

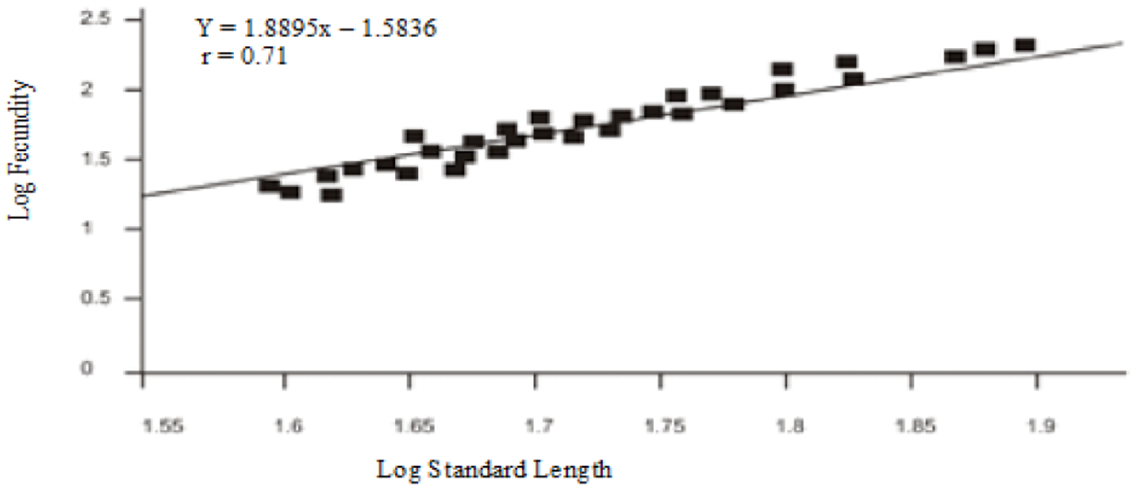

Figure 5a: Log Fecundity – Log Standard length relationship of *Papyrocranus afer* from Lekki Lagoon

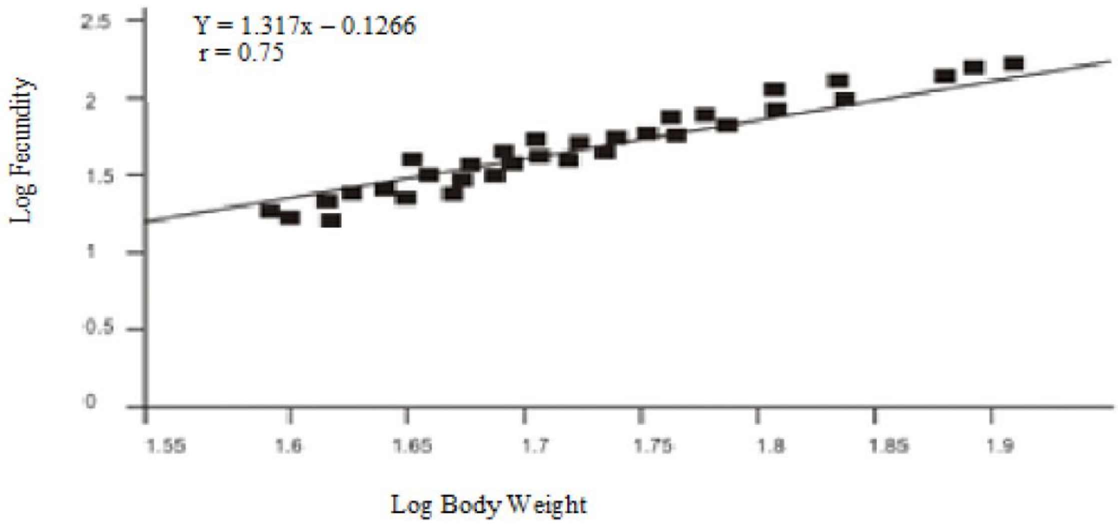

Figure 5b: Log Fecundity – Log Body weight relationship of *Papyrocranus afer* from Lekki Lagoon

Egg diameter ranged between 2.8 – 4.1 mm with mean 3.6 ± 0.3 mm from ripe/mature stages.

GSI was higher in females 0.14 – 0.68 % with mean value of 0.44 ± 0.14 % than in males 0.1 – 0.5 % with mean value of 0.22 – 0.01 %. Monthly variation in GSI of both males and females are illustrated in Figure 5. The lowest mean values was obtained in January for both male and female 0.4 ± 0.1 and 0.6 ± 1.2 respectively; while the highest values of 0.9 ± 0.2 and 1.7 ± 1.1% were recorded in July for both male and females respectively. Variations in the gonad weight and GSI of the female fish reached to the peak during July indicating maturity of ovary and definite spawning season.

Logarithmic transformation of GSI-weight relationships of male and female (Figures 6a and b) indicated a strong relation for female (r=0.97), and weak (r=0.13) for female. There was a significant difference in the mean weight sizes of male (214.83 ± 196.82) and female (520.91 ± 201.82); t (533) = -13.1, p<0.001.

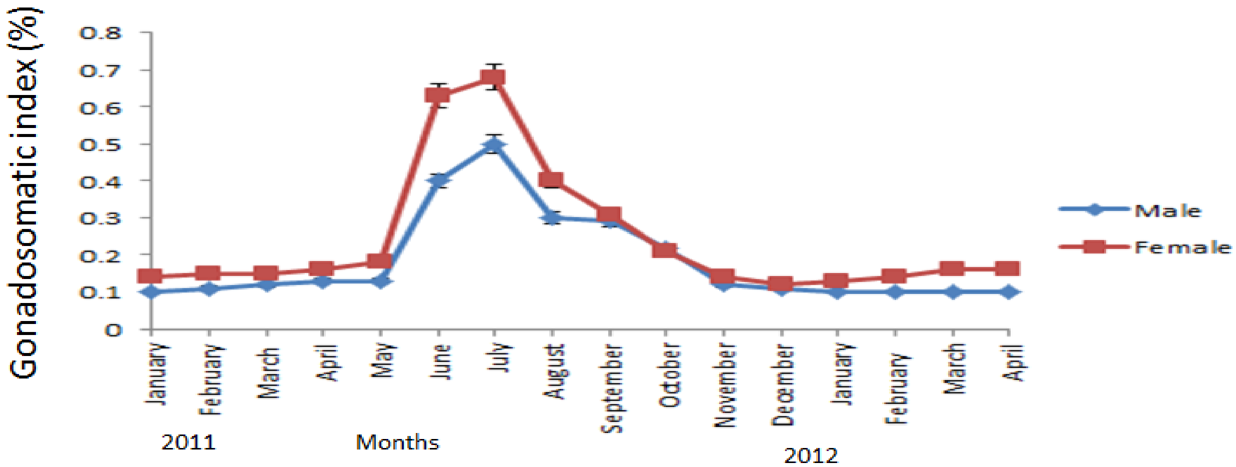

Figure 7: Log fecundity – Log body parameters relationship of *Papyrocranus afer* from Lekki Lagoon

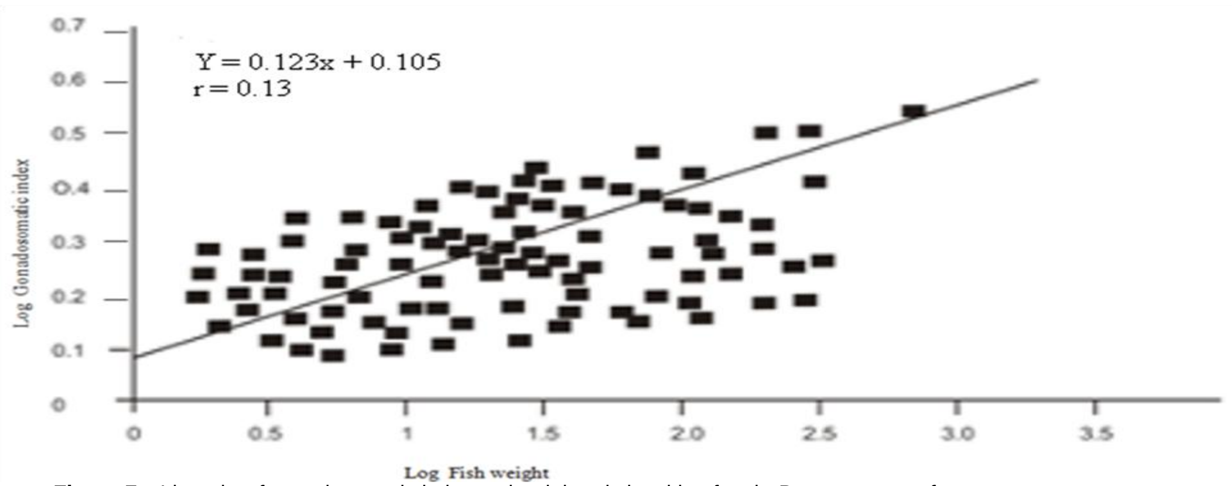

Figure 7a: Linearity of gonadosomatic index and weight relationship of male *Papyrocranus afer*

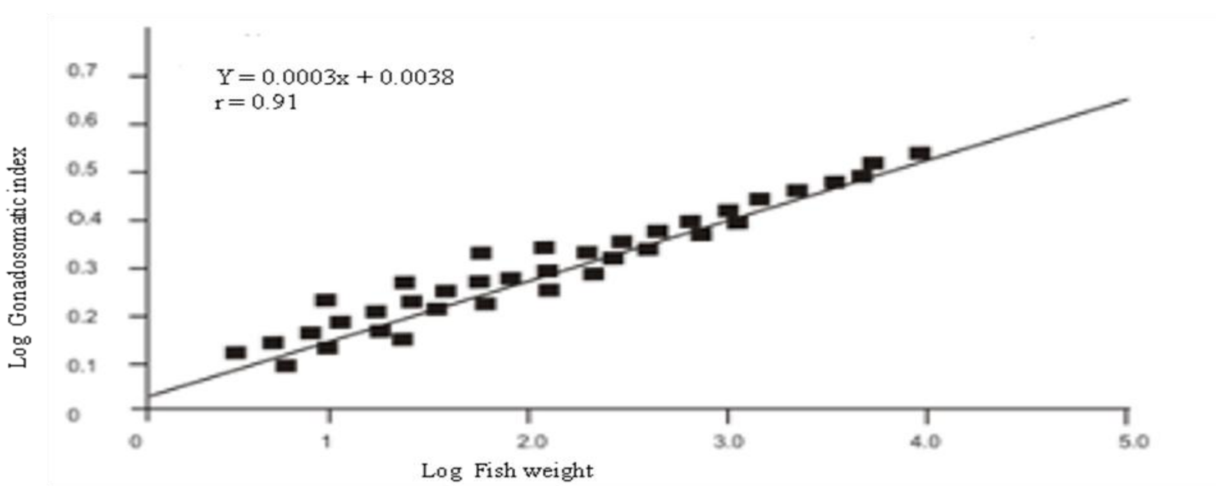

Figure 7a: Linearity of gonadosomatic index and weight relationship of female *Papyrocranus afer*

DISCUSSION

Overfishing could occur in two ways: growth overfishing which is excessive harvesting of younger individuals in a population, resulting in lower mean sizes of caught fish, and hence lower biomass harvested; and recruitment overfishing which is excessive harvesting of the reproductive spawning stock, such that reproduction and recruitment decline (Cushing, 1977). In the present study, recorded wide difference between the most frequently caught specimens and the sizes at maturity indicated growth overfishing for the stock of *Papyrocranus afer* from Lekki Lagoon. This could be attributed to increasing collection of the juvenile for aquaria by artisanal fishermen to argument their income. Consequently, restriction of harvest of early life stages of *P. afer* is imperative for sustenance of this fish stock at Lekki Lagoon. Sustainable fisheries require sufficient annual recruitment to balance population losses to harvest (Welcomme, 2001).Minimum legal length (MLL) describes the minimum sizes of fish that can be harvested by law from a population; this size is usually defined as the length at which 50% of the population is sexually mature (Koob, 2011). The results of this study can be directly used to support this management measure. Nigeria currently does not have a MLL for fishes in water bodies including *P. afer*, but implementing it would assist in preventing further decline in population of this species from growth over-fishing. Fishing gear restrictions are also closely related to the use of MLL as ornamental fisheries strategy.

Maximum sizes recorded for *Papyrocranus afer* were 66.9 cm and 75.1 cm standard lengths in males and females respectively. This differed from the maximum size = 80 cm recorded for this species, unsexed on FishBase (Froese and Pauly, 2019) which could be attributed to environmental or genetic factors; combination of these two categorical factors. Estimated values of L_{m50} in male (49.1 cm) and female (53.4 cm) *P. afer* corresponded to more than 70 % of the maximum attained length in this study. Agosthiho *et al.* (1991) showed that tropical fish species attained sexual maturation around 40 to 50 % of their maximum attainable length. This generalization is in contrast with observation in the present study. *P. afer* exhibited higher values than 70 % in both sexes. This indicated late maturity for *P.a efer* in Lekki Lagoon. Rossoni *et al.* (2010) reported that the 50 % length corresponding to maximum attainable length in both sexes for discus fish, *Symphysodon aequifasciatus*, from Purus River (Amazonas, Brazil) corresponded to more than 60 % of the maximum attained length. Fish species with delayed maturity are more likely to be at risked of over-exploitation than species with early maturity (Parent and Schriml, 1995, Musick, 1999). Late maturity is a characteristic often associated with increased risk of over-exploitation (Sadovy and Cheung, 2003) Duarte *et al.* (2011) reported that males are smaller than females in most fish species; this strategy is associated with greater reproductive in put by larger females for greater oocytes production as larger females have a larger peritoneal cavity and thus can lay more eggs. Both fertility and the number of fertilized eggs increase with body size, a general pattern in teleosts (Gross and Sargent, 1985). Similarly, in the present study, males are generally smaller than females.

In dioecious species, sex ratios are expected to be close to unity. Rossoni *et al*. (2010) reported that differential growth of males and females may influence sex ratio structure, resulting in the predominance of one sex or the other throughout development stages. Higher percentages of male to female fish in favour of the males during spawning seasons have been noted in African catfish, *Chrysichthys walkeri* (Ikusemiju, 1975)Adebayo (2013) observed the dominance of male to females in sompat grunt, *Pomadysys jubelini,* from the Lagos coast, Nigeria.

Several authors have reported that differential growth, influence of gear selectivity, exploitation rate, coupled with ecological factors such as temperature could cause deviation from the expected sex ratio of 1:1 (male-female) (Bohlen *et al.,* 2008; Olurin and Savage, 2011; Imam *et al.,* 2012). Male biased sex-ratio has been reported in certain Cypriniformes species *Cobitis elongatoides* and *Sabanejewia balcanica* by Bohleen *et al.* (2008). To my knowledge, there is dearth of data on sex ratio of notopterids including *Papyrocranus afer*. However, Controlled breeding in captivity was done at sex ratio 1: 2 (female: male) for Asian feather back knife fish, *Notopterus notopterus* at Bandladesh Fisheries Research Institute, Mymensingh, Bandladesh, India (Jahan *et al.,* 2015). Yanwirsal *et al.* (2017) at the Humboldt University, Germany successfully attempted the controlled breeding of *Notopterus notopterus* at both sex ratios 2:1 and 1:1 (female/male).

Low fecundity of *Papyrocranus afer* may have resulted from low visceral cavity for holding the eggs and relatively large size of the eggs although not reported in this study. The egg number and size are both dependent on the visceral capacity of the female female (Beldade *et al.,* 2012). The number of eggs produced by fish was shown by Nikolsky (1963) to be related to the degree of parental care.In the notopterids, *Notopterus notopterus* and *Chitala chitala* perfom parental care usually done by the males (Johan *et al.,* 2015; Yanwirsal *et al.,* 2017). Fishes that produced millions of eggs (ocean sunfish, *Mola mol;* Cod, *Gadus* species) exhibited no parental care (Fryer and Iles, 1972). On the other hand, females that produced one or a few hundred eggs (sticklebacks, *Gasterosteus* species) often exhibited a form of parental care (Fryer and Iles, 1972; Fagade and Adebisi, 1979; Beldade *et al.,* 2012; Mohamed *et al.,* 2013). Lagler *et al.*

(1977) reported that as a rule, the fecundity of fish is inversely related to the degree of parental care.

Fecundity was positive and linearly correlated with fish size indicating that both body weight and total length are reliable indicators of the fish's potential egg production. Fagade and Adebisi (1979) reported that in *Chrysichthys nigrodigitatus*, body weight was a more reliable indicator than length for estimation of the fish's potential egg production; Fawole (2002) found that in *Mormyrus rume,* the correlation between fecundity and body weight (r= 0.56) was higher than that between fecundity and standard length (r= 0.47). Females of *P. afer* in Lekki Lagoon were heavier than males of the same length. This could be attributed to additional weight gain in ovaries of female especially during the breeding season.

Specimens in this study were found to have mature gonads during wet season months, indicating annual breeding pattern. The recorded GSI suggested the spawning period of *P. afer* coincided with the rainy season with peak in July. GSI was higher in the rainy season than in the dry season, further confirming that breeding and spawning in this species takes place during the rainy season. This result was in line with those of Svensonn (1933) reporting that spawning occurred in *P. afer* during the rainy season in the swamps of Gambia. Also *Notopterus notopterus*, in India mainly was found to spawn during the rainy season (Parameswaran and Sinha, 1966). Jahan *et al.* (2015) reported that in all tropical species of osteoglossidae, reproduction is indeed related to the high and low water seasons. Males had lower gonadosomatic indices than females of corresponding gonad stage of development. This is in line with the assertion that gonadosomatic indices are generally higher in females than males for African species (Munro *et at.,* 1990); due to additional weight gain of ovaries in the breeding period as a result of accumulation of yolk in the eggs and also due to uptake of fluid by ripe oocyte (Shinkafi and Ipinjolu, 2012).

This paper reports for the first time sexual dimorphism in *Papyrocranus afer* based on the structure of the genital papillae. Other important results to take into consideration were the late sexual maturity, male-biased sex ratio, low fecundity as well as seasonal onset and distribution of annual spawning as the reproductive attributes of *Papyrocranus afer* in Lekki Lagoon. This information could serve as baseline information for management of this species in Lekki Lagoon.

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