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Some aspects of the reproductive biology of tilapia black-chinned, *Sarotherodon melanotheron* (Rüppel, 1852) in relation to macrophyte abundance in three marginal lagoons, Southeast of Côte d'Ivoire

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Abstract

Sarotherodon melanotheron is a cichlid species found in abundance in most of estuaries and lagoons of West African. In recent decades, these fish are facing problems with pollution, wetland degradation, macrophyte proliferation and fishing pressure. This study examined some aspects of its reproductive biology and the possible influence of macrophyte proliferation on the reproductive parameters. A total of 1054 fish were collected monthly with cast nets, gillnets, traps, harpoons and hawks between 2017 and 2019. The temperature, transparency, pH, total dissolved solids (TDS), conductivity and dissolved oxygen (DO) were recorded in *situ*. Water samples were taken, stored in polyethylene bottles (500 mL) and kept at a temperature below 4° C for further determination of nutrients (ammonium-nitrogen NH₄⁺, nitrate NO₃⁻, nitrite NO₂⁻ and phosphate PO₄³⁻). The overall sex ratio was in favour of males in all lagoons. The length at first sexual maturity (L₅₀) was approximately equal between males and females from the same lagoon. However, it increased as the macrophyte abundance decreased from Ono to Hebe lagoons or at the end of the dry season in Hebe lagoon. The presence of females with ripe gonads throughout the year gives evidence of multiple spawning and attested that this species reproduces in all seasons. The absolute fecundity was higher in Ono lagoon, indicating a better food supply due to the high abundance of macrophytes.

Keywords: Aquatic plants; Cichlid; fecundity; sex ratio; spawning period; West Africa

1. Introduction

Studies of fish reproduction is crucial for the sustainable development of aquaculture. Reproductive strategies vary greatly between fish, particularly among cichlid species [1,2]. Some fish populations have developed adaptation strategies to cope unusually stressful environmental conditions [3,4,5]. The overall pattern of reproduction is common to all species and covers a range of life history traits including the age and size at first maturity, gonadal development, fecundity and oocyte sizes. However, fish can develop alternative reproductive tactics to respond to fluctuation in the environment. Both overall and alternative strategies are adaptive as they ensure the survival of the species in specific environmental conditions [6].

The black-chinned tilapia *Sarotherodon melanotheron* (Rüppel, 1852) is an estuarine species of West African lagoons and estuaries that can reproduce in a wide salinity range [7]. The aspects of its biology have been reported by Eyeson [8,9], Koné and Teugels [10] and Arizi et al [11]. However, most of these studies were conducted in brackish water and no study was carried out in marginal lagoons which are strongly affected by anthropogenic activities and macrophytes infestation. Since more than two decades, the surface of these ecosystems includes a wide variety of macrophytes [12]. The combination of these factors has led to the deterioration of the water quality, with respect to dissolved oxygen and

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nutriment concentrations. This study aims to assess the effect of macrophyte abundance on sex ratio, size at first maturity, fecundity and spawning season of *S. melanotheron* in Ono, Kodjoboue and Hebe lagoons.

2. Materials and methods

2.1. Study area

Ono lagoon (5°22'22"N and 3°33'53"W), Kodjoboue lagoon (5°14'11 "N and 3°35'9" W) and Hebe lagoon (5°12'14" N and 3°33'15" W) are three small lagoons of 400 ha, 423 ha and 244 ha, respectively in the Southeast of Côte d'Ivoire (Figure 1). The percentage of macrophyte abundance was determined using satellite images by the University Centre for Research and Application in Remote Sensing of the University Félix Houphouët Boigny, Côte d'Ivoire. Although these lagoons are invaded by macrophytes, Ono lagoon is almost completely invaded (60-70%) by a wide variety of habitat types such as native (*Echinochloa Pyramidalis, Pistia stratiotes, Nymphaea lotus*) and non-native (*Eichhornia crassipes, Hydrilla verticillata, Salvinia molesta*) plants, reducing its exploitable surface to 162 ha. In the other lagoons, only the banks are covered by macrophytes, with a coverage rate of 20% for Kodjoboue lagoon and less than 2% for Hebe lagoon. Permanently connected to the Comoe River, these lagoons have an equatorial climate, including two rainy seasons (April–July and October–November) and two dry seasons (December–March and August–September). The permanent linkage with the Comoe River produces typical freshwater characteristics of these lagoons.





2.2. Environmental parameters sampling and analysis methods

The temperature, transparency, pH, total dissolved solids (TDS), conductivity and dissolved oxygen (DO) were recorded in *situ*. Water samples were taken stored in polyethylene bottles (500 mL) and kept at a temperature below 4°C for further determination of ammonium-nitrogen (NH₄⁺; mg/L), nitrate (NO₃⁻; mg/L), nitrite (NO₂⁻; mg/L) and phosphate (PO₄³⁻; mg/L). The samples were filtered through Whatman GF/C fibreglass filters and concentrations were determined using a spectrophotometer Model HACH DR 6000.

2.3. Data collection and laboratory procedure

Fish were monthly sampled from 2015 to 2016 using cast nets, gillnets, traps, harpoons and hawks. The specimens were transported in iced box to the laboratory of the Department of Aquatic Living Resources of the Oceanological Research Centre, Abidjan. Each individual was measured to the nearest 1 mm (standard length [SL]), weighed to the nearest 0.01

g using an electronic balance. Fish was then dissected, and the gonads were removed and weighed to the nearest 0.001 g. A large collection of specimens was collected during the study period, enabling a maturity scale to be developed for each sex. Gonads were macroscopically assessed using the scale of Arthington and Milton [13]. However, to reduce the chance of error in correctly identifying individual stages, gonads were classified as either immature (juvenile and inactive stages) or mature (maturing, ripe and spent stages) (Table 1).

Table 1 Macroscopic scale in male and female Sarotherodon melanotheron caught in Ono, Kodjoboue and Hebe lagoonsfrom 2017 to 2019 (Arthington and Milton [13]).

Stages of gonad development	Description
I (immature)	Gonads not fully formed but present as two tiny transparent threads of tissue. Sex difficult to distinguish macroscopically.
II (Inactive)	Ovary slightly yellowish, oocytes macroscopically distinguishable. Testis pale, thin and strap-like.
III (Maturing)	Ovary greatly enlarged, oocytes readily visible and bright yellow. Testis broadened, distended and cream in colour.
IV (Ripe)	Oocytes are of maximum size, readily extruded from female under slight abdominal pressure. Testis swollen to maximum size.
V (Spent)	Ovary partly empty with irregular distribution of oocyte diameters. Some mature and developing oocytes detectable. Testis flaccid but still at maximum size.

Average size at first maturity (L_{50}) was defined as the size at which 50% of individuals in the population were matures during the reproduction period. Fish were grouped into 10 mm size classes and 20 to 30 individuals were examined within each. The L_{50} was estimated from the proportion of mature individuals in each 10 cm size class, using the logistic function of Ghorbel et al [14]:

$$P = \frac{e^{(\alpha + \beta SL)}}{1 + e^{(\alpha + \beta x SL)}}$$

where P = percentage of mature fish, SL = standard length, and α and β = coefficients.

This equation can be transformed into a logarithmic form as follows:

$$\frac{ln \ P}{1\text{-}P} = \alpha + \beta SL$$

The value of L₅₀ was estimated from the negative ratio $-\alpha/\beta$ by substituting P = 0.5 in the above equation.

The gonadosomatic (GSI) index, which represents the gonad weight expressed as a percentage of the wet body weight, Gonad weight (g) = 100

was estimated using the following formula: $GSI = \frac{Gonad \text{ weight } (g)}{Eviscerated \text{ weight } (g)} \times 100$

The absolute fecundity is the number of oocytes likely to be released at the next spawning. In *S. melanotheron*, the oocyte distribution in the ovary is multimodal and ripe females (stage 4) are more likely to lay oocytes with the highest modal diameter. Therefore, only the stage 4 from females were used to estimate the fecundity. After weighing the ovaries, sub-samples were taken according to the size of the ovaries and all oocytes of each sample were counted manually because the oocytes of this species are large and easy to manipulate. A mean fecundity from all samples was calculated using a "direct summation" procedure [15].

$$d = \sqrt{(d_1 \, x \, d_2)}$$

2.4. Statistical analyses

The Shapiro-Wilk normality test for homoscedasticity were applied to the data, to determine whether the assumptions of the parametric or nonparametric analyses for abiotic parameters, sex ratio and GSI were satisfied. A chi square (χ^2) test was applied to test the significant differences between the sex ratio of females and males. The analysis of variance (ANOVA) was used to test the spatial variation of environmental parameters and the monthly variations of mean GSI values for each separate sex. Tukey's HSD multiple contrasts test was used to determine significant differences at the 0.05 level.

3. Results

3.1. Environmental variables

The environmental variables of Ono, Kodjoboue and Hebe lagoons were presented in Table 2. Significant variation in water parameters was observed among the sampling lagoons (ANOVA test, p < 0.05), except for nitrite and ammonium-nitrogen. However, no significant different (ANOVA test, p > 0.05) was observed between Kodjoboue and Hebe lagoons for all parameters. For the parameters such as temperature, pH, DO, conductivity and TDS, the values were significantly higher in Hebe lagoon whereas the values were lower for the temperature, pH and DO in Ono lagoon and for conductivity and TDS in Kodjoboue lagoon. Values of transparency, nitrate and phosphate were lower in Hebe lagoon and higher in Ono lagoon.

Parameters	Ono lagoon	Kodjoboue lagoon	Hebe lagoon
Temperature (°C)	27.15 ± 1,55 ^a	29.71 ± 1.56 ^b	29.91 ± 1.37 ^b
Dissolved oxygen (mg/L)	2.27 ± 0.83^{a}	5.79 ± 0.96 ^b	6.20 ± 0.52^{b}
рН	6.30 ± 0.48^{a}	6.27 ± 0.56^{a}	6.65 ± 0.43^{b}
Conductivity (µS/cm)	17.09 ± 5.91 ^a	12.97 ± 6.52^{a}	34.89 ± 22.45 ^b
TDS (mg/L)	8.06 ± 1.52 ^a	5.96 ± 3.20 ^a	15.87 ± 3.16 ^b
Transparency (m)	1.57 ± 0.30 ^b	1.19 ± 0.21^{a}	0.69 ± 0.32^{a}
Nitrate (mg/L)	3.07 ± 0.91 ^b	2.29 ± 0.65 ^a	1.75 ± 0.55 ^a
Nitrite (mg/L)	0.25 ± 0.40^{a}	0.22 ± 0.47^{a}	0.15 ± 0.33^{a}
Ammonium-nitrogen (mg/L)	0.07 ± 0.03^{a}	0.05 ± 0.03^{a}	0.06 ± 0.03^{a}
Phosphate (mg/L)	0.47 ± 0.23^{b}	0.26 ± 0.14^{a}	0.30 ± 0.11^{a}

Table 2 Average values (mean ± SD) of the physical and chemical parameters in Ono, Kodjoboue and Hebe lagoons between September 2017 to August 2019.

3.2. Sex-ratio

A total of 341 specimens (189 males and 152 females), 311 (174 males and 137 females) and 402 (241 males and 161 females) were collected in Ono, Kodjoboue and Hebe lagoons, respectively (Table 3). The overall sex ratio was in favour of males in Ono (1: 0.80; $\chi^2 = 4.01$; p < 0.05), Kodjoboue lagoon (1: 0.79; $\chi^2 = 4.40$; p < 0.05) and Hebe (1: 0.67; $\chi^2 = 15.92$; p < 0.05) lagoons. Seasonal variations were also observed, with females being numerous in April–August (Ono lagoon) and February–April (Kodjoboue lagoon) and males in the remaining months. In Hebe lagoon, the situation was totally different as males were preponderant throughout the year.

	Ono lagoon				Kodjoboue lagoon			Hebe lagoon				
Month	Male (n)	Female (n)	Ratio M: F	χ ²	Male (n)	Female (n)	Ratio M: F	χ ²	Male (n)	Female (n)	Ratio M: F	χ ²
J	25	12	1:0.48	4.57*	15	10	1:0.67	1.00	25	12	1:0.48	4.57*
F	18	14	1:0.78	0.50	11	18	1:1.64	1.69	22	15	1:0.68	1.32
М	15	8	1:0.53	2.14	14	16	1:1.14	0.13	18	13	1:0.72	0.81
А	14	15	1:1.07	0.04	14	15	1:1.07	0.03	18	17	1:0.94	0.03
М	16	17	1:1.06	0.04	10	9	1:0.90	0.05	18	11	1:0.61	1.70
J	12	13	1:1.08	0.04	15	13	1:0.87	0.14	22	13	1:0.59	2.31
J	10	17	1:1.70	1.82	18	14	1:0.78	0.50	21	15	1:0.71	1.00
А	15	19	1:1.27	0.48	15	12	1:0.80	0.33	19	14	1:0.74	0.76
S	15	10	1:0.67	1.00	17	11	1:0.65	1.29	18	14	1:0.78	0.50
0	13	9	1:0.69	0.73	17	10	1:0.59	1.81	18	16	1:0.89	0.12
N	13	8	1:0.62	1.19	15	6	1:0.40	3.86*	17	8	1:0.47	3.24
D	23	10	1:0.43	5.12*	13	3	1:0.23	6.25*	25	13	1:0.52	3.79
Total	189	152	1:0.80	4.01*	174	137	1:0.79	4.40*	241	161	1:0.67	15.92*

Table 3 Monthly proportion (%) of male and female Sarotherodon melanotheron caught in Ono, Kodjoboue and Hebelagoons from 2017 to 2019.

3.3. Size at sexual maturity

The length at first sexual maturity was 147.5 mm, SL for females and 148.1 mm for males in Ono lagoon, 148.3 mm for females and 149.7 mm for males in Kodjoboue lagoon and 154,8 mm for females and 155.0 mm for males in Hebe lagoon (Figure 2). The difference between sexes in the same lagoon was not significant (p > 0.05). However, individuals of Hebe lagoon reached sexual maturity at large sizes than those of the other lagoons. The least matured male and female in the population were 84.0 mm for both sexes in Ono and Kodjoboue lagoons whereas the smallest male and female with ripe gonads measured 97.0 mm and 80.0 mm, respectively in Hebe lagoon.





Figure 2 Size at first sexual maturity (L₅₀) for male and female *Sarotherodon melanotheron* caught in Ono, Kodjoboue and Hebe lagoons from 2017 to 2019.

3.4. Spawning periodicity

The variation in mean GSI values of both sexes followed nearly the same pattern in the three lagoons, with significant differences in male and female between months (ANOVA, p < 0.05). The GSI values of females and males (Figure 3) increased after July and peaked in October (Ono and Kodjoboue lagoons) and in September (Hebe lagoon) followed by a decrease progressively throughout the flood period until the beginning of the dry season in December. A second increase in GSI occurred from January to March, reaching a peak in February–March and then declining from April to June. In Hebe lagoon, the first peak occurred earlier at the end of the small dry season (August).



Figure 3 Monthly mean gonadosomatic indices (GSI) of male and female *Sarotherodon melanotheron* from Ono, Kodjoboue and Hebe lagoons. Error bars represent ± SD.

All stages appeared throughout the sampling period in all lagoons (Figure 4). The proportion of immature stages (I and II) was higher in December–January and May–July for Ono lagoon, in December–January and March–August for Kodjoboue lagoon and in December and June–July for Hebe lagoon. The proportions of maturity stages indicated a high prevalence of sexual stages III, IV and V for both males and females from February to April and from August to November (Ono lagoon), from February to April, June and from September to November (Kodjoboue lagoon) and from January to March and from August to December (Hebe lagoon). The maturing stage (III) peaked one month before the peak of pre-spawning stage (IV) in both sexes. The highest proportions of pre-spawning (stage IV) were obtained in September–October and February–March (Ono and Kodjoboue lagoons) and in August–October and January–March (Hebe lagoon). The spent stages (stage V) of Ono lagoon reached the maximum percentages in November–December, those of Kodjoboue lagoon in October–November and those of Hebe lagoon in October–November and May.



Figure 4 Percentages of maturity stages for females and males of *Sarotherodon melanotheron* from Ono, Kodjoboue and Hebe lagoons. Stl: immature; StII: inactive; StIII: maturing; StIV: ripe; StV: spent.

The percentage of individuals with mature gonads (stages III, IV and V) and mean monthly GSI were correlated for both males and females, attesting that the peak spawning periods were from August to November and from February to April.

Most of spent females occurred in October (Ono and Kodjoboue lagoons) and in September (Hebe lagoons), corresponding to the peak of maturing females in September and August, respectively.

3.5. Effects of macrophyte abundance on reproductive cycle

The variation of macrophyte abundance present the same trend in both lagoons, with values showed two peaks (February–March and July–September) followed by a relatively decrease from April to June and from February to April (Figure 3). The peak of reproduction occurred mainly in October and September when macrophyte abundance was relatively higher. The Table 4 showed no significant correlation between GSI and macrophyte abundance regardless of sex in both lagoons.

Table 4 Summary of correlation coefficients (r) between macrophyte abundance and GSI in Ono Kodjoboue and Hebelagoons from 2017 to 2019.

	Sexes	n	R ²	r	p value
Ono lagoon	Female	12	0.0560	0.2366	0.4590
	Male	12	0.0350	0.1872	0.5601
Kodjoboue lagoon	Female	12	0.0295	0.1719	0.5932
	Male	12	0.0023	0.0484	0.8813
Hebe lagoon	Female	12	0.0676	0.2601	0.4143
	Male	12	0.0070	0.0837	0.7960

3.6. Fecundity

The absolute fecundity ranged from 107-320 oocytes (216 ± 49 oocytes), from 103-298 oocytes (211 ± 42 oocytes) and from 105-295 oocytes (201 ± 56 oocytes) corresponding to fish sizes of 110-226 mm, 110-213 mm and 110 to 206 mm in Ono, Kodjoboue and Hebe lagoons, respectively. The relationship between the standard length and fecundity was linear with a stronger correlation for the relationship in Ono (r = 0.78), Kodjoboue (r = 0.70) and Hebe (r = 0.67) lagoons (Figure 5).





Figure 5 Relation between fecundity and standard length of *Sarotherodon melanotheron* sampled in Ono, Kodjoboue and Hebe lagoons from 2017 to 2019.

4. Discussion

The overall sex ratio (M: F) of *S. melanotheron* is found in different numbers with obvious deviation from the expected ratio (1: 1) where the number of males exceeded that of females in all lagoons. Previous authors have noted an overall male predominance in *S. melanotheron* [16, 11]. Several hypotheses, including reproduction, displacement for foraging, the practice of mouth brooding, differential growth and mortality by sex could explain this result [17]. Fryer and Iles [2] pointed out that, in African water bodies, it is common in the Cichlid populations that males dominate because of fast growth, so that they are more susceptible to exploitation than females. In addition, due to their mouth brooding and egg-protecting behaviour, males are more sedentary in the fishing ground, which would make them more vulnerable to gears.

There were no significant variations in size at first maturity between the males and females of *S. melanotheron* in the same lagoon, although females showed slightly smaller size at L_{50} than males. According to Bagenal [18], females invest more energy on gonadal development, resulting in their maturing at smaller sizes. Furthermore, individuals of Hebe lagoon reached sexual maturity later than those of Ono and Kodjoboue lagoons. Eyeson [8] identified the temperature as the parameter affecting sexual maturation and spawning in tilapia. This contrasts with the current research results as the temperature did not vary significantly throughout the year. We rather observed the increase of the size at first sexual maturity with increasing dissolved oxygen from Ono lagoon to Hebe lagoon. This sexual size also seems to be negatively correlated with the infestation degree of macrophytes which decreased according to this same gradient. The

observed reduction of the size at maturity in Ono lagoon is due to the strong human activities which have degraded its biotic and abiotic environment over the past two decades. Russell et al [19] and Waithaka et al [20] found that in unstable environments, temperature, dissolved oxygen, habitat size, and water level can fluctuate significantly; which is advantageous for tilapias that exhibit r- life history traits characterized by early maturation. Ono lagoon includes a wide variety of habitat types such as native and non-native plants whose decomposition process absorbs oxygen from the water, which can accentuate anoxic conditions. On the other hand, this lagoon has been affected for several years by industrial and domestic pollutant discharges, and natural eutrophication caused by increased nutrient concentrations. The decomposition process of dead macrophytes absorbs oxygen from water and may therefore induce anoxic conditions. Fish subjected to such pressures would have exhibited physiological adaptations related to growth or reproduction namely early sexual maturity and dwarfism. Similar observations have been reported in several species of fish [21, 5] In contrast, the absence of macrophytes in Hebe lagoon associated to the increased freshwater inputs from precipitation may help to dilute pollutants and therefore relieving fish from these stress factors.

With regards to the monthly changes of the GSI and the males and females with ripe gonads in all lagoons, *S. melanotheron* displayed multiple reproduction with several offspring cohorts during the same spawning season. This breeding pattern is typical to most teleost fishes and cichlids in particular [22]. The breeding activity started in dry season and became intense in wet season (rainy and flood seasons) or at the end of the dry season (September, Hebe lagoon), a pattern commonly documented for fish species inhabiting large rivers and wetlands in Africa [23, 24]. This species therefore reproduces in the rainy seasons when the environmental and trophic conditions remain favourable for several months to ensure the development and survival of its offspring. Mahoney and Gibson [25] and Rey et al [26] reported that this period coincides with the occurrence of high rainfall events that in turn cause harpacticoid copepods, phytoplankton and miscellaneous zooplankton to bloom which are the major food sources for black-chinned tilapia. This cycle contrasts with the observation of continuous reproduction reported in lagoons [21] and lakes [27], but is similar to the marked cycle described under a more sub-tropical climate [16]. The highest spawning activity during the rainy season can be attributed to the high increase in the dilution of contaminants by precipitation and not to a variation of temperature. Indeed, our study did not show any direct or indirect influence of this parameter on the reproduction of *S. melanotheron* in the areas studied, as evidenced by the absence of significant seasonal variation.

The range of fecundity in this study is lower than the fecundity of 107–580 oocytes found by Fagade [28] for *S. melanotheron* in Lagos lagoon, probably due to the effect of surrounding human activities and the abundance of macrophytes in these lagoons. Within a given species, fecundity may vary as a result of different adaptations to environmental habitat [29] and differential availability in food resources [30]. This higher fecundity in Ono lagoon appears to be related to a better food supply due to the high abundance of macrophytes as this species is omnivorous with a phytophagous tendency [31, 32]. Rennie and Jackson [33] pointed out that, macrophytes provide habitat complexity and breeding areas, as well as being substrata for periphyton and sites of abundant food production for many aquatic animals. For Siddiqui et al [34], the variation in fecundity may be attributed to differential abundance of food within members of population.

5. Conclusion

Although the seasonality of the reproduction does not appear to be affected by temperature, this study showed that dissolved oxygen, nutrient concentrations and macrophyte infestation lead to changes in reproductive parameters. The main modification concerns the change in the size at maturity and fecundity, with a reduction of fish sizes when dissolved oxygen increases and macrophyte infestation decreases. These results have significant implication on understanding the impact of environmental variability on timing and intensity of the reproduction cycle for *S. melanotheron*.

Compliance with ethical standards

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Disclosure of conflict of interest

I declare that I have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Statement of ethical approval

Fish sampling methodology was approved via the Ministry in charge of Animal and fishery resources in Abidjan, Côte d'Ivoire.

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