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## Variations of reproduction in Mascarene Grass Frog in Banco National Park, Côte d'Ivoire

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### Abstract

The reproduction of *Ptychadena mascareniensis* (Ptychadenidae) was studied in rainy and dry seasons in three habitats (Bay, Fish Farm and Filtisac) in Banco National Park (Ivory Coast). After weighing and dissection of different specimens, male (75) and female (67) gonads are weighed. The eggs of female gonad were counted. The sizes at first sexual maturity for males and females are respectively 41.80 mm and 50.40 mm in Bay, 44.25 mm and 51.10 mm in Fish Farm and 43.75 mm and 54.75 mm at Filtisac. The sex ratio is in favor of females in all sites except for the rainy season at Filtisac where it's in favor of males (1.17 / 1). The spatial variations of the gonado somatic index (GSI) in females recorded are 6.30%, 7.54% and 6.54% respectively at Bay, Fish Farm and Filtisac. Regarding males, the median values of GSI (0.48%; 0.19% and 0.22% observed in Bay, Fish Farm and Filtisac respectively) did not vary significantly. Regarding the fertility, the average values of absolute fertility in Bay, Fish Farm and Filtisac are 834, 997 and 644 oocytes respectively. It was concluded that there was a significant difference in the variability of absolute fertility is observed between the fish farm and Filtisac. Regarding the seasons, the difference is significant only between absolute fertility in the dry season in the Bay and in the rainy season at the fish farm.

**Keywords:** Reproduction, *Ptychadena mascareniensis*, First sexual maturity, Banco national park, Côte d'Ivoire

### 1. Introduction

The spatio-temporal distribution and the reproduction of amphibians are greatly influenced by the availability of water in the habitat. These animals are aquatic, terrestrial, fossorial or arborea [1-5]. They are found in habitats ranging from savannas [6, 7] to humid primary forests [8] to the regions of high altitudes [9-12]. Amphibians do not live in salt environments. However, some accommodate themselves to brackish water. Mascarene Grass Frog is a species of savannah that is also encountered in anthropised forests. In Ivory Coast, particularly, this frog is found in all savannas and degraded habitats of forest areas [13-14-15-16-17-8]. In the Banco National Park, vestige of primary forest situated in the heart of Abidjan, the economic capital of Côte d'Ivoire, the data about the ecology of amphibians [8, 18-24] are recent. Very little data on the reproduction of *Ptychadena mascareniensis* is in our possession. Yet this big frog is an important source of animal protein in this country. To ensure the sustainability of the resource, the mastery of its breeding is necessary. In Côte d'Ivoire, particularly in the Banco National Park, only one publication [22] relates its diet. Regarding its reproduction, the results available in this ecosystem refer only to its pace of activity songs [20]. This study was, therefore, focused mainly in evaluation of the spatial and temporal variation of the reproduction of *Ptychadena mascareniensis* in Ivory Coast.

### 2. Materials and Methods

#### 2.1. Study environment

The present study has been carried in the Banco National Park (Figure 1). His description, location and characteristics were presented by [21]. These same authors have described in detail the different sites which house shelter the specimens of *Ptychadena mascareniensis*

#### 2.2. Sampling and analysis

*Ptychadena mascareniensis* were captured with a dip net during the day and night from

February to June 2015. After identification basis on work of [25] and measuring the Snout-Vent Length (SVL) to the nearest millimeter with a caliper, the frog is anesthetized with chlorobutanol and its gonad is collected. It is then weighed and kept in pill dispensers with 70% ethanol for laboratory studies. The counting of oocytes was done with a mechanical counter.

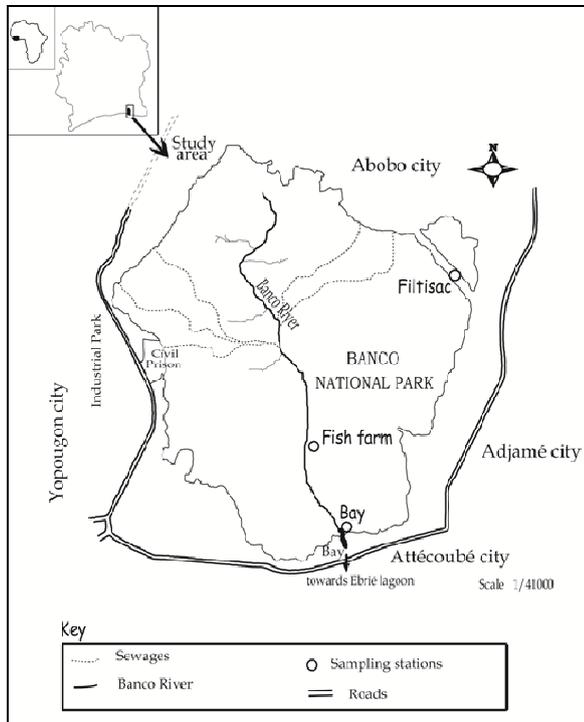


Fig 1: Presentation of the Banco National Park and study sites [21]

### 2.3. Sex ratio (SR)

The proportion of males or females (sex-ratio) in a population of species may be expressed as a percentage in the number of males or females compared to the sampled population:

$$SR = (M \times 100) / (M+F) \text{ or } SR = (F \times 100) / (M+F)$$

M = number of males; F = number of females.

### 2.4. Size of first sexual maturity

The size of first sexual maturity (or size of first reproduction or L50) is a standard average length from which 50% of individuals can reproduce [26, 27]. It is defined from the size classes based on Sturge's formula in [28].

Interval of classes = (maximal size – minimal size) / (number of classes)

Number of classes  $N = 1 + 3.3 \log n$

with n is the number of individuals for the sample considered.

This method which is widely used in fish reproduction study [26, 27, 29] takes account only mature individuals and their cumulative percentages. To get it, you use the different median of the size classes in abscissa and the cumulative percentage of mature individuals per size classes on ordinate. The abscissa point corresponding to the projection of 50% of

individuals on the curve gives the mean size at first maturity. From these curves, we can distinguish three categories of individuals according to the size [30].

- L0: size below which no individual is mature;
- L50: size from which there is as many mature than immature individuals;
- L100: size at which all individuals are mature.

### 2.5. Gonadosomatic Index (GSI)

Gonadosomatic index (GSI) or "Gonadosomatic Report (RGS) is defined as the ratio of gonad weight to body weight percentage.

$$RGS = (W_g \times 100) / (W)$$

Wg = gonad weight; W = weight of the frog.

We considered the eviscerated weight of the frog which presents the advantage of eliminating the gonad weight, the weight of digestive tract and its contents. A very higher ratio indicates a advanced state of gonads maturation. In contrast, a low ratio is in favor of sexual rest.

### 2.6. The absolute fecundity

The gonads are removed from pill dispensers and deposited on absorbent paper to remove the alcohol. They were stripped of their envelope. These oocytes were separated using pliers. The absolute fecundity is obtained by counting all the oocytes effectively present in ovary using a manual particle counter.

we used the Kruskal-Wallis and Mann-Whitney non-parametric tests to compare the reproduction with respect to sites, seasons and fertility. We referred to STATISTICA version 7.1 for the non-parametric tests.

## 3. Results

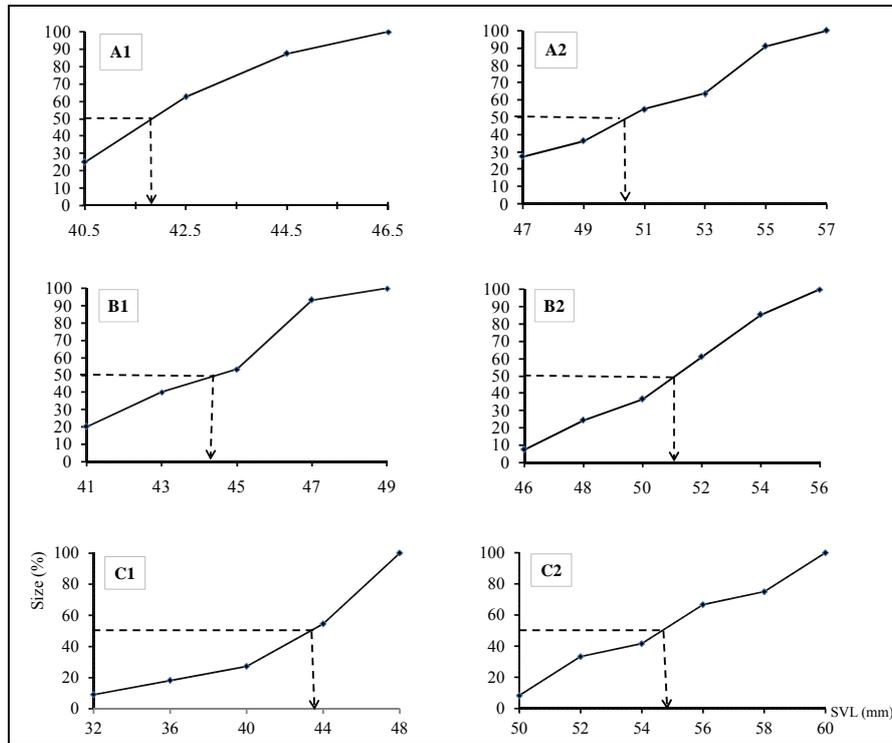
### 3.1 Size of first sexual maturity

The sizes of first sexual maturity studied in males and females of *Ptychadena mascareniensis* at Bay, Fish farm and Filtisac in the Banco National Park are illustrated in figure 2.

At Bay, the sizes of first sexual maturity calculated (L50) is 41.80 mm for males (Figure 2A) against 50.40 mm for females (Figure 2B). The smallest mature individual measure was 40.5 mm and 47 mm respectively for males and females. All specimens reach sexual maturity at 46.5 mm in males and 57 mm in females.

At the Fish Farm, L50 of mature females is 51.10 mm (Figure 2B2) against 44.25 mm for males (Figure 2B1). The respective sizes of smaller mature individuals in this habitat are 46 mm and 41 mm. All mature individuals have a size greater than 56 mm in females and 49 mm males.

At Filtisac, the L50 is 43.75 mm for males (Figure 2C1) and 54.75 mm for females (Figure 2C2). The smallest mature individuals respectively measure 32 mm and 50 mm. All specimens are mature at 60 mm for females and 48 mm for males.

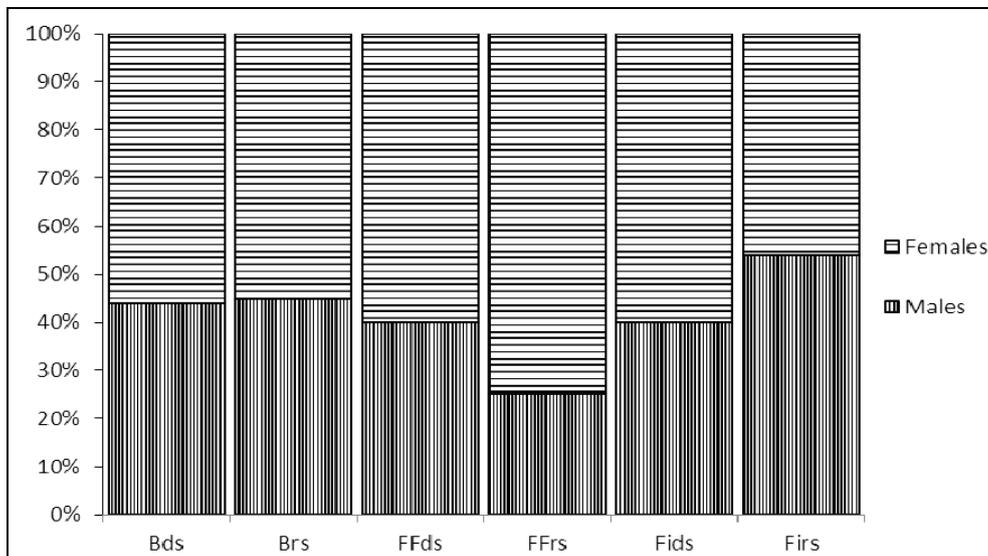


**Fig 2:** Variation in sizes at first sexual maturity in males (1) and females (2) of *Ptychadena mascareniensis* at Bay (A), Fish farm (B) and Filtisac (C) in the Banco National Park: SVL = snout vent length.

**3.2. Sex ratio**

Figure 3 shows the variation in sex ratio in the dry and rainy seasons at the bay, fish farm and Filtisac. The sex ratio is in

favor of females in all sites except in the rainy season at Filtisac where it's in favor of males (1.17 / 1).



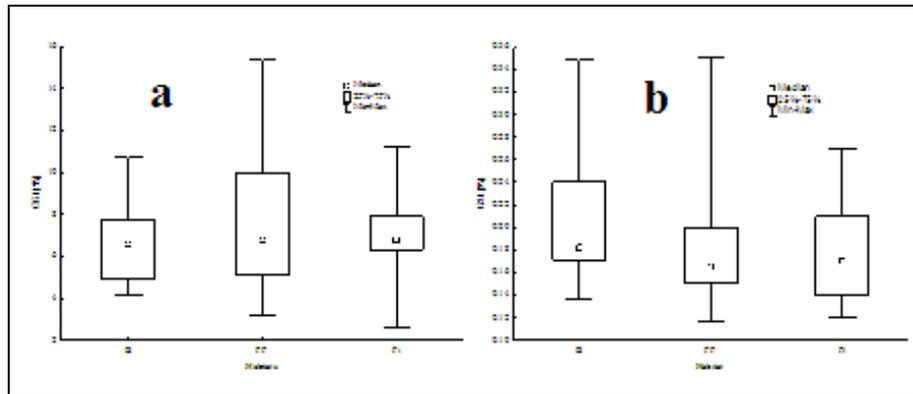
**Fig 3:** Seasonal variation in sex ratio of *Ptychadena mascareniensis* at the Bay (B), fish farm (FF) and Filtisac (Fi) : ds = dry season; rs = rainy season

**3.3. Gonado-somatic index (GSI)**

**3.3.1. Spatial variation of the gonado somatic index (GSI)**

Figure 4 illustrates the spatial variation of GSI in male and female specimens of *Ptychadena mascareniensis* at Bay, Fish farm and Filtisac. In females (Figure 3a), the medians of GSI recorded are 6.30%, 7.54% and 6.54% respectively at Bay,

Fish Farm and Filtisac. The Kruskal-Wallis test shows no significant difference between these values in the three stations ( $P = 0.4910$ ). Regarding males (Figure 4b), the median values of GSI (0.48%; 0.19% and 0.22% respectively) observed in Bay, Fish Farm and Filtisac) did not vary significantly (Kruskal Wallis test,  $P = 0, 5812$ ).

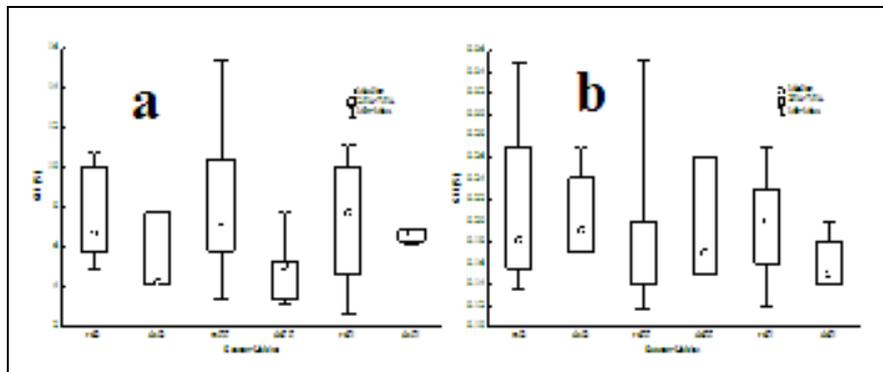


**Fig 4:** Change in GSI of female (a) and male (b) specimens of *Ptychadena mascareniensis* at bay (B), fish farm (FF) and Filtisac (Fi).

**3.3.2. Seasonal variation of the gonado-somatic index (GSI)**

The changes in GSI of females (Fig. 5a) and males (Fig. 5b) of *Ptychadena mascareniensis* specimens were reported in Figure 5. This setting varies with the seasons in both sexes. In

females, the highest value of the median (8.2%) was observed during the rainy season in Filtisac and the lowest in the Bay (4.3%) in the dry season.



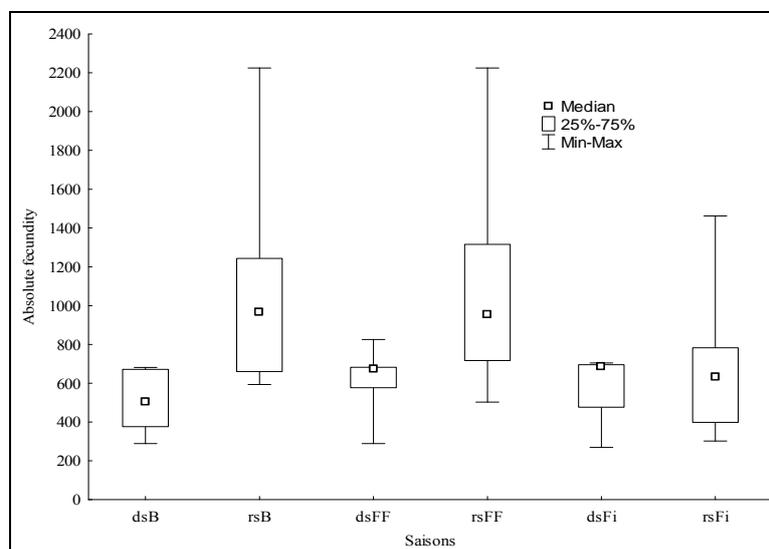
**Fig 5:** Seasonal variation of the gonado-somatic index (GSI) in male (a) and female (b) of *Ptychadena mascareniensis* at bay (B), fish farm (FF) and Filtisac (Fi) : rs = reany season ; ds = dry season.

However, differences between habitats in the different seasons do not show any significant differences (Kruskal Wallis,  $p = 0.1439$ ).

For males, the highest median (0.23%) and the lowest median (0.17%) were recorded in Filtisac in rainy season and dry season respectively. These differences still are not significant (Kruskal Wallis,  $P = 0.6941$ ).

**3.3.3. Variation in fertility**

The study of fertility was conducted on 67 ovaries from three sampling sites. The average values of absolute fertility in Bay, Fish Farm and Filtisac are 834, 997 and 644 oocytes respectively



**Fig 6:** Seasonal variation of the absolute fertility at Bay (B), Fish farm (FF) and Filtisac (Fi) in Banco National Park : ds = dry season ; rs = rainy season.

Kruskal-Wallis's test indicates a significant difference in the variability of absolute fertility between the fish farm and Filtisac ( $p = 0.0325$ ). Figure 6 illustrates the seasonal variations of absolute fertility in the three habitats.

The highest median values were observed during the rainy season in Bay (1109 oocytes) and fish farm (1062). The lower values were observed in the dry season in Bay (504 oocytes). The difference is significant only between absolute fertility in the dry season in the Bay and in the rainy season at the fish farm ( $p = 0.0258$ , Mann Withney's test).

#### 4. Discussion

The analysis of the results obtained shows that the size at first maturity is higher in females. Similarly, the size of the smallest mature individual captured is greater in females than in males. The males would then be more precocious than females. However, Our values were lower than those of [25]. This author reported 43 mm in males and females of *Ptychadena mascareniensis* in the Tai reserve. The difference between these results could be explained by the environment of this frog. In the Banco National Park, this edible anoure would be seriously threatened. Thus, to ensure their survival, the specimens would adapt by replicating itself early.

According to the seasonal and spatial variation of the RGS, our results show that the peaks were obtained in females in the three habitats during the rainy season. This could be attributed to the low temperatures and high humidities offered by the rainy season. These major differences are in favor of a strong sexual activity among frogs. This has already been observed by [31] for *Hoplobatrachus occipitalis* in the Banco National Park. Furthermore, [32] reported for *Hyperilius nasutus* that low temperatures and high relative humidity of the rainy seasons were a stimulating factor for oocyte maturation. As for the sex ratio which is in favor of females in the rainy season, it would be linked to territoriality that exists in frogs. In fact, the song of the male during the breeding period, while also attracts females, its beacon territory against the approach of other males. This strategy that limits the competitions in the conquest of the female by the male could explain the high number of females captured during this period. The analysis of the relative fecundity in the three habitats reveals the higher value at the Fish Farm than in the other two sites. This result could be attributed to the low temperatures and high relative humidity of that habitat that are more conducive to the breeding of frogs. This agrees with the data from [25] in Comoé National Park. Indeed, this author points out that frogs are fond of very moist habitats for reproduction.

#### 5. Acknowledgements

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