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Biological characteristics of the reproduction of the land crab *Cardisoma armatum* (Herklots, 1851) in Ehotile National Park (Cote d'Ivoire)

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Abstract

Aim: *Cardiosoma armatum* is a Land crab that is widely exploited and consumed in Côte d'Ivoire. This study aims to highlight some reproductive parameters of this species in the Iles Ehotilé National Park in relation to sex ratio, size at first sexual maturity, spawning season and fecundity. Methods: Sampling was carried out monthly from January to December 2018 using dip nets, trap boxes, hole fishing and collection. Results: Reproductive parameters of *C. armatum* were examined on 930 specimens ranging in size from 61.35 to 80.86 mm carapace width and eviscerated weights from 96 to 160 g. The overall sex ratio of 1.14:1, favoured males. The size at first sexual maturity was 64.60 mm carapace width in females and 64.27 mm in males. The monthly gonado-somatic index and the macroscopically determined gonad stages indicated that *C. armatum* has two favourable spawning periods in the PNIE, from June to October, with spawning in August, and from November to April, with spawning in February. The mean monthly varied between 219958 and 567848 oocytes in females. The results of this study suggest that *C. armatum* has very good reproductive parameters and can be used as a breeding crab.

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Introduction

The land crab Cardisoma armatum is a decapod crustacean in the family Gecarcinidae. It is a tropical species known from the islands of Cape Verde and Senegal to Angola, including Fernando Poo and the islands of Sao Tomé in the Gulf of Guinea (Manning and Holthuis, 1981). C. armatum, commonly known as the hairy crab, is widely distributed in Ivorian towns such as Assinie, Aboisso, Adiaké, Bassam, Bonoua, Abidjan, Dabou, Sassandra, Fresco and San Pedro (D'almeida et al., 2014). The species is coastal and mangrove-dwelling. The combined analysis of the gut coefficient and diet indicates that C. armatum is an omnivorous species with a tendency to be an invertivore (N'zi and Coulibaly, 2021). The proteinrich meat of this crab is highly valued by riverine communities (d'Almeida et al., 2014) and forms an integral part of their diet. Fishing for C. armatum, a species of commercial interest, is intense, with adult females being the main target. This often abusive exploitation has a negative impact on the renewal of the exploited population (FAO, 2004).

To prevent the collapse of C. armatum stocks, a rational, integrated management policy is needed to ensure the sustainable use of this resource. One of the scientific underpinnings of this policy is an understanding of the reproductive biology of the species. Knowledge of reproductive parameters is essential for rational fisheries management (Offem et al., 2008). In Côte d'Ivoire, research on C. armatum has focused on a number of ecological aspects, such as sex ratio and length-weight relationships (Etchian et al., 2016), general characteristics and embryonic development of eggs (D'almeida et al., 2014). Other aspects are still poorly understood. For example, the stages of sexual maturity make it possible to characterise the different states of the ovaries and testes during their development; the sex ratio provides information on the reproductive behaviour of the species; the size at first sexual maturity of males and females provides information on the proportion of the stock capable of ensuring the renewal of the species; the somatic gonad ratio determines the state of sexual activity of the species

during the course of the year; the RHS, which defines the energy status required for gonad maturation; the condition factor (K), which makes it possible to estimate seasonal changes in body weight under the influence of environmental and/or physiological factors; fecundity, which provides information on population dynamics; the coefficient of variation of oocytes, which determines the egg-laving characteristics of the species, etc. The aim of this study is to investigate and provide information on the reproductive parameters of C. armatum in the Iles Ehotilé National Park, to contribute to the development of a sustainable management policy and to obtain the data necessary for the captive breeding of this species in Côte d'Ivoire.

Material and methods

Study environment

The Ehotilé National Parck (PNIE) (Fig. 1) is located in the south-east of Côte d'Ivoire in the department of Adiaké, between longitudes 3°16'43' and 3°18'52' west and latitudes 5°9'45' and 5°11'12' north. It comprises 6 islands: Assoko (327.5 ha), Baloubaté (75 ha), Méha (45 ha), Nyamouin (47.5 ha), Elouamin (22.5 ha) and the sacred island of Bosson-Assoun (32.5 ha) (Lauginie, 2007). All these islands are surrounded by the Aby lagoon and separated from each other by arms of lagoon water giving the park a total area of 722 ha (OIPR, 2018). Based on the diversity of the biotopes, 10 sampling stations (Assoko 1, Assoko 2, Balouaté 1, Balouaté 2, Elouamin 1, Elouamin 2, Méha 1, Méha 2, Niamouin 1, Niamouin 2) were surveyed.

Sampling and laboratory analysis

Crabs were sampled between January and December 2018 using dip nets, trap boxes, hole fishing and collection. In the laboratory, each specimen was measured using a MITUTOYO GENERAL model digital caliper with an accuracy of 0.01 mm and weighed using a Denver SI-4002 type balance with an accuracy of 0.01g and a capacity of 400 g. Each specimen was then dissected, the hepatopancreas and gonads removed and weighed to the nearest gram.



Fig. 1. Crab sampling stations in Ehotilé National Park (PNIE), between January and December 2018. A = Assoko, B = Balouaté, E = Elouamin, M = Méha, N = Nyamouin.

Mathematical expressions data

Sex ratio

The sex ratio (SR) has been defined as the ratio of males to females (Kartas and Quignard, 1984). The formula is as follows: SR=M/F; where M = number of males and F = number of females.

Size at first sexual maturity

Size at first sexual maturity (LC50) was estimated by adjusting the percentage of mature individuals by the logistic function of a non-linear regression according to Ghorbel *et al.* (1996). The equation is: : $P = \frac{1}{1 + e^{-(\alpha + \beta L \zeta)}}$; with P = percentage of mature individuals and LC = carapace width; α and β = constants. The LC₅₀ deduced from the previous equation is defined as follows: : $LC_{30} = \frac{-\alpha}{\beta}$

Gonadosomatic and hepathosomatic ratios

The gonadosomatic (RGS) and hepathosomatic (RHS) indices, which respectively represent the weight of the gonads and the hepathopancreas, expressed as a percentage of the wet body weight of the crab, have been estimated respectively (Stephen, 1988).

RGS =
$$\frac{\text{Weight of the gonad }(\textbf{g})}{\text{Total crab mass }(\textbf{g})} \times 100$$
 et
RHS = $\frac{\text{Weight of hepatopancreas }(\textbf{g})}{\text{Total weight }(\textbf{g}) \text{ of crab}} \times 100$

Condition factor

Condition factor (K) was calculated using the formula defined by Kartas and Quignard (1984). $K = \frac{\text{Crab weight (g)}}{[\text{Carapacewidth (cm)}]^2} \times 100$

Fecundity and oocyte diameter

Fecundity is the number of mature oocytes present in the ovary immediately prior to oviposition (Bagenal, 1973). Fecundity was calculated from oocytes at stage 4 of sexual maturity. Absolute fecundity (Fa) and relative fecundity (Fr) were calculated using the following formulae: $\mathbf{Fa} = \operatorname{Ni} \mathbf{x} \frac{M_{\mathfrak{x}}}{M_{f}}$ et $\mathbf{Fr} = \frac{Ni}{M_{i}}$; Ni = number of oocytes present in a fraction of the ovary; Mt = total mass of the ovary (g); Mf = mass of the fraction of the ovary (g); Nt = total number of oocytes

produced; Mt = total mass of the crab.

Oocyte diameter and coefficient of variation

Oocyte diameter was obtained from measurements of 90 oocytes. The formula for the coefficient of variation of oocyte diameters defined according to Ouattara (2000) is as follows: $Cv = \frac{S}{m_{DQ}} X 100$; s = standard deviation of mean oocyte diameter and mDO = mean oocyte diameter. The classification according to Ouattara (2000) is as follows : Cv < 2% : Very homogeneous structure ; $2\% \le Cv < 30\%$: Homogeneous structure and $Cv \ge 30\%$: Heterogeneous structure.

Statistical processing

A Mann-Whitney test was used to compare Gonado-Somatic Ratio, Hepato-Somatic Ratio and condition factor between crab sexes and between hydrological seasons. The Chi-squared Test was used to compare sex proportions by month and season. Statistical differences were estimated at the 5% significance level. All statistical analyses were performed using Statistica 7.1 version.

Results

Sex ratio

930 specimens of C. armatum were examined, divided between 496 males (53.33%) and 434 females (46.67%). The overall sex ratio determined was 1.14:1 (M : F) and favoured males. The chi-square test showed no significant difference ($\chi^2 = 0.24$; p > 0.05) in the sex ratio between males and females. The monthly variation in sex ratio (Fig. 2) shows that the sex ratio favours females in January (57.58%), February (64.29%), March (61.90%), June (60%), July (62.5%) and August (55.56%). Males dominate in April (65.38%), May (73.08%), September (59.46%), October (56.76%), November (58%) and December (78.26%). The monthly trend in the sex ratio of C. armatum showed a significant variation in this parameter in February ($\chi^2 = 5.27$; p < 0.05), March $(\chi^2 = 3.86; p < 0.05)$ and July $(\chi^2 = 3.86; p < 0.05)$ in females and April ($\chi^2 = 6.06$; p < 0.05), May ($\chi^2 =$ 14.45; p < 0.05) and December ($\chi^2 = 21.66$; p < 0.05) in males. Seasonal variation showed that during the

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dry season, the highest proportion of males was recorded in December (78.26%) and that of females in February (64.29%). During the rainy season, the highest proportion of males was recorded in May (73.08%) and that of females in July (62.5%). The Chi-square test revealed a significant difference between the two seasons ($\chi^2 = 0.01$; p < 0.05).

Size at first sexual maturity

The size at first sexual maturity of *C. armatum* specimens was 64.60 mm carapace width in females (Fig. 3) and 64.27 mm in males (Fig. 4). The size of the smallest mature individual observed in females and males was 47.38 mm and 47.78 mm carapace width, respectively. No significant difference was observed between the size at first sexual maturity and the size of the smallest mature individual in males and females (Mann-Whitney U test, p < 0.05).

Monthly variations in the stages of sexual maturity

The monthly frequency of occurrence of the different stages of sexual maturity in females is shown in Fig. 5. Stage 1 maturity was observed throughout the sampling period except in March and August. Stage 2 sexual maturity was observed in March (15.38%), June (8.33%), July (23.08%) and October (6.25%). Stage 3 ovaries were found throughout the sampling period except in August and December. Stage 4 ovaries were observed from January to March, with a peak in February (63.16%), and from June to December, with a higher percentage in August (100%). Stage 5 individuals were observed in October (6.25%), January (21.05%), February (5.26%) and March (23.08%).

For males (Fig. 6), stage 1 sexual maturity was observed throughout the year except in February and March, with a higher proportion in July (55.56%). Individuals at stage 2 sexual maturity were observed throughout the year, with the exception of January and March. The highest percentage of mature individuals was observed in December (27.78%). Stage 3 sexual maturity was observed throughout the year, with a peak in February (30%). Specimens at stage 4 were observed throughout the year, with the exception of July, and the peak of maturity was recorded in March (75%). Stage 5 specimens were observed from January to May, then in July and from September to December, with a higher percentage in September (31.82%).

3.4 Gonado-somatic ratio

Monthly variations in the Gonado-somatic ratio (RGS) are shown in Fig. 7. Monitoring the average monthly GSR of C. armatum females shows that this parameter varies between 0.21% in April and 2.59% in August. There are two peaks. The first was recorded in February (2.41%) and the second in August (2.59%). The lowest RGS was recorded in April (0.21%). The monitoring of the average monthly RGS of males (Fig. 7) of C. armatum shows that this parameter varies from 0.03% in September to 0.45% in July. The first peak was recorded in February (0.29%) and the second in July (0.45%). Overall, the first peaks in male and female RGS coincide with February. For the second peaks, the male peak coincided with July and the female peak with August. The mean RGS of the females $(1.61\% \pm 0.76)$ is higher than that of the males ($0.18\% \pm 0.12$). Statistical analysis using the Mann-Whitney U test showed a significant difference between the RGS of males and females (p < 0.05).

Seasonal variation of RGS shows that in females, it is higher (2.15 \pm 0.41) in the dry season than in the rainy season (1.21% \pm 0.72). In males, the RGS is higher (0.19 \pm 0.13) in the rainy season than in the dry season (0.16 \pm 0.10). For both sexes combined, RGS was higher in the dry season (1.16 \pm 1.08) than in rainy season (0.70 \pm 0.72). The Mann-Whitney U test showed no significant difference in RGS between seasons (p > 0.05).

Hepatosomatic ratio

Monthly variation of the hepatosomatic ratio (RHS) of males and females is shown in Fig. 8. The female RHS reached its maximum in July (10.17%) and its minimum in August (3.64%). For males, the RHS reached its maximum in June (9.85%) and its minimum in May (6.39%). Analysis of the mean RHS

of females (7.41% \pm 1.82) showed that it was lower than that of males (7.96% \pm 1.11). The Mann-Whitney U test showed no significant difference between the RHS of male and female C. armatum (p > 0.05).

Seasonal variation in RHS showed that that of females was high (7.74% \pm 1.79) in the rainy season and low (6.93% \pm 1.96) in the dry season. On the other hand, that of males is high (8.15% \pm 0.82) in the dry season and low (7.83% \pm 1.32) in the wet season. For both sexes combined, the RHS was higher in the wet season (7.78% \pm 1.51) than in the dry season (7.54% \pm 1.56). The Mann-Whitney U test showed no significant difference between seasons (p > 0.05).

Condition factor

The condition factor (K) of *C. armatum* fluctuates between 37.79 and 44.22 with an average value of 42.06 \pm 3.98 in females (Fig. 9). It varied between 39.18 and 45.35 with an average value of 42.95 \pm 3.66 in males. Analysis of the monthly variation curves shows that in *C. armatum*, overweight is low in September and November in males and females respectively. It is highest in March for both sexes. Mean values for this parameter are higher in males than in females. Mann-Whitney U test showed no significant difference (p > 0.05) in overweight between the sexes in *C. armatum*.

Seasonal variation in the condition factor showed that it was higher for males and females in the rainy season than in the dry season and for both sexes combined. The Mann-Whitney U test showed no significant seasonal variation (p > 0.05).

Fecundity and oocyte diameter

The estimated absolute fecundity of *C. armatum* females in the PNIE ranged from 219958 to 567848 oocytes with a mean value of 331273 ± 162038 for specimens with a carapace width between 61.35 and 80.86 mm and an eviscerated weight between 96 and 160 g. The relative fecundity ranged from a minimum of 1476 oocytes to a maximum of 3542 oocytes per unit of body weight. The mean fertility was 2274 \pm 937 oocytes per gram of body weight. Oocyte

diameter ranged from 0.7 to 2 mm with a mean of 1.10 ± 0.16 mm. Analysis of oocyte size based on the coefficient of variation (Cv = 39,58) showed that the oocytes had a heterogeneous structure.

Discussion

Cardisoma armatum crabs caught in the PNIE show a dominance of males over females, with an overall sex ratio of 1.14:1. In Nigeria, similar results were observed by Olalekan *et al.* (2015) in the same species with a sex ratio of 1.12. The opposite result was observed by Gbemisola *et al.* (2005) in the same species with a sex ratio of 1.18 in favour of females. This difference in sex ratio shows that it can vary within the same species and according to the characteristics of the environment.

The monthly observation of the sex ratio in our study showed an uneven distribution of sex ratios. The monthly variation in the sex ratio could be attributed to periodic movements in search of food and/or areas suitable for reproduction.

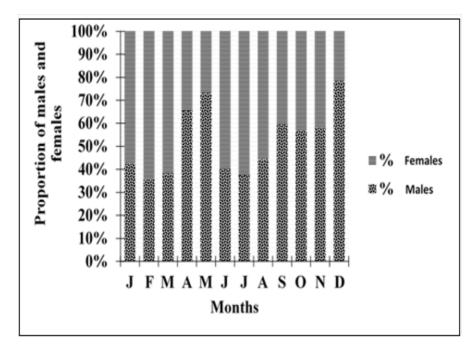


Fig. 2. Monthly variation in the proportion of males and females of *Cardisoma armatum* sampled in PNIE (Côte d'Ivoire) from January to December 2018.

The seasonal variation of the sex ratio showed that during the dry season, the highest proportion of males was observed in December (78.26%), followed by females in February (64.29%). In the rainy season, the highest proportion of males was recorded in May (73.08%), followed by females in July (62.5%). The high density of males before females in all seasons could be explained by the search for a sexual partner. Indeed, in the months two months after each male abundance, there is a peak in RGS for females. As for the abundance of females in February and July, this could be due to the search for food to help the eggs mature. In fact, February coincides with a peak and July precedes the second, larger peak in female RGS.

general, the proportion of females decreases progressively after each peak in the RGS. On the other hand, the proportion of males increases during the same period. According to King and Etim (2004), the sex ratio is influenced by several factors such as reproduction, foraging, differential growth and natural mortality by sex. The reduction in the number of females over time is thought to be due to their displacement from mangrove areas to specific habitats for larval release after gonad maturation. Larval release requires higher salinity than in mangrove areas (Stephen, 1990). In the PNIE, males reach sexual maturity before females. The size at first

The monthly variation in the sex ratio shows that, in

sexual maturity is 64.60 mm and 64.27 mm carapace width for females and males respectively. These results show that C. armatum reaches first sexual maturity later in both males and females in the PNIE compared to crabs of the same species recorded by Goussanou *et al.* (2018) in Benin. This author recorded a first sexual maturity size of 58 mm in males and 60 mm in females.

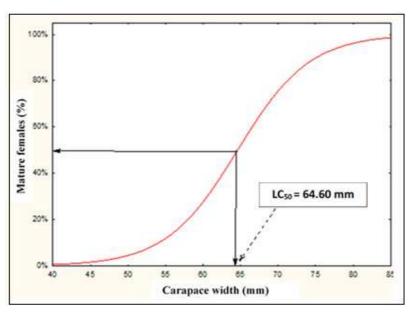


Fig. 3. First sexual maturity size (LC₅₀) of *Cardisoma armatum* females caught in PNIE, Côte d'Ivoire, January-December 2018.

According to Ouattara *et al* (2008), this difference in size at first maturity could be explained by a lack of available food and/or over-exploitation of stocks. Other authors, such as Montchowui *et al* (2007), attribute the spatial variation in size at first maturity to environmental conditions. The PNIE is one of the protected areas where no human activity is allowed. This protection of the park allows the crabs to escape exploitation, feed better and grow to large sizes before reproducing.

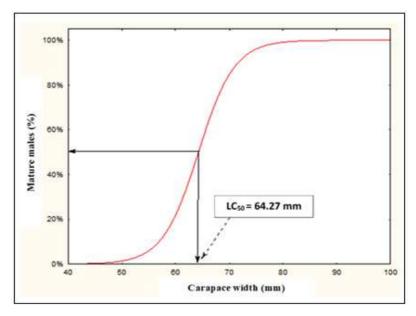


Fig. 4. Size at first sexual maturity (LC₅₀) of male *Cardisoma armatum* caught in PNIE, Côte d'Ivoire, January-December 2018.

The RGS gives an indication of the development and activity of the gonads (Etim *et al.*, 1989). The average monthly RGS measured in C. armatum in the PNIE shows that that of females is higher than that of males, with a significant difference (U-test, p < 0.05). This higher RGS value in females is thought to be due to the high number and large size of these oocytes (Paugy *et al.*, 2006).

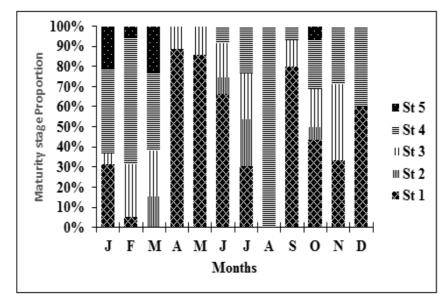


Fig. 5. Monthly variation in sexual maturity stages of *C. armatum* females sampled in PNIE (Côte d'Ivoire) from January to December 2018. St = stage.

The monthly trend in RGS shows two main peaks, one in February and a larger one in August. The percentage of mature females in the 4 preporte stage was observed in two periods, from January to March with the highest value in February (63.16%) and from June to December with a higher percentage in August (100%). The high proportions of females in stage 4 prepontia in February and August (dry season) corroborate those of Stephen (1990) and Heasman *et al.* (1985), who obtained Scylla serrata females in the prepontia stage during the warm season in New Caledonia and Australia, respectively.

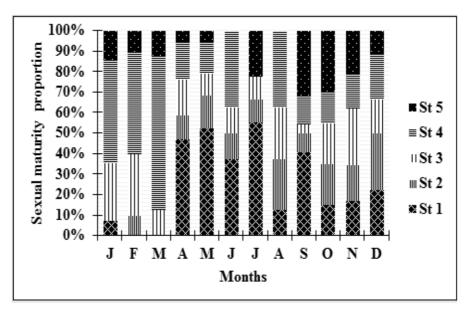


Fig. 6. Monthly variation in sexual maturity stages of *C. armatum* males sampled in PNIE (Côte d'Ivoire) from January to December 2018. St = stage.

Individuals in stage 5, corresponding to oviposition, were observed in October (6.25%), January (21.05%), February (5.26%) and March (23.08%). The combined analysis of the peaks in the average monthly RGS of the females and the high proportions of mature females in pre-spawning stage 4 and spawning stage 5 indicates two periods that are favourable for the reproduction of *C. armatum* females. The first period is from June to October with an oviposition period from August to October and a

second favourable period for reproduction is from November to March with an oviposition period from January to March. According to Islam and Bhuiyan (1982), the movements and onset of spawning appear to be dependent on the lunar cycle and linked to specific salinity conditions. In general, reproduction occurs more or less before a period when environmental factors are most favourable for juvenile survival and hence for the survival of the species (Koné, 2000).

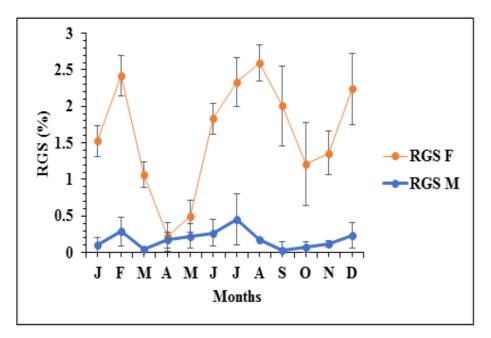


Fig. 7. Monthly variations in the gonadosomatic ratio (RGS) of females (F) and males (M) of *C. armatum* sampled in the PNIE (Côte d'Ivoire) from January to December 2018.

The higher proportion of individuals at stage 5 of spawning in March (23.08%) could be explained by the arrival of the rains, which would be an important environmental factor to enable the larvae to feed well after their release into the aquatic environment.

According to Rotimi *et al.* (2012), the eggs laid are carried by the abdomen of the female for about two weeks before migrating to the sea for release, as larval release requires higher salinity than in mangrove areas. In fact, rain and run-off water provide a greater amount of food for aquatic organisms during the rainy season (Castillo-Rivera, 2013). Juvenile crabs released into the sea during the rainy season may benefit from the availability of food and shelter in submerged areas.

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In the present study, biological dormancy occurs during the rainy season in April. This period is characterised by the lowest RGS value (0.21%). This result corroborates that of Stephen (1990), who recorded genital dormancy during the cool season (austral winter) in the mangrove crab Scylla serrata.

Periodic variations in RHS and condition factor (K) provide specific information on the organs that accumulate lipid reserves and their role in mobilising reserves during gonad maturation (Zouari, 2010). These organs may be the hepatopancreas or the muscles, or both. In the present study, the condition factor (K) and the RHS developed in the same order of magnitude. This observation shows that the organ that accumulates lipid reserves for the maturation of *C. armatum* oocytes may be the hepatopancreas.

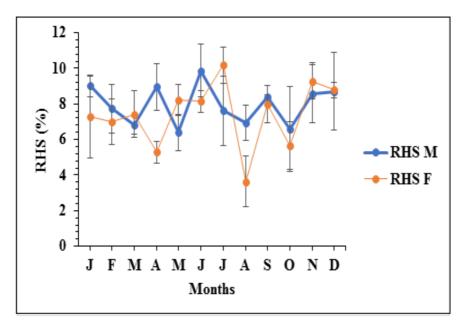


Fig. 8. Monthly variation of the hepato-somatic ratio (RHS) of females (F) and males (M) of *C. armatum* sampled in the PNIE (Côte d'Ivoire) from January to December 2018.

For *C. armatum* in the PNIE, the mean condition factor was 42.95 ± 3.66 for males and 42.06 ± 3.98 for females. The mean value of this parameter is higher than that obtained in males (40.33 ± 8.49) , but lower than that recorded in females (47.00 ± 6.97) of *C. armatum* in the Comoé River (Côte

d'Ivoire) by Etchian *et al.* (2016). The recorded condition factor is also higher than that observed for C. armatum in Nigeria by Olalekan *et al.* (2015) (30.7). These observations show that the value of the condition factor can vary between individuals of the same species and from one environment to another.

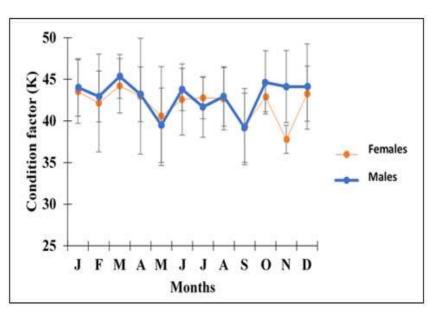


Fig. 9. Monthly variation of condition factor (K) in males (M) and females (F) of *C. armatum* sampled in the PNIE (Ivory Coast) from January to December 2018.

According to Berté (2009), the difference between condition factors of populations may be related to the quantity and/or quality of food available in different environments. The seasonal variation indicates that the condition factor for both sexes combined is higher in the wet season than in the dry season. The Mann-

Whitney U test showed no significant seasonal variation (p > 0.05), indicating that *C. armatum* is well adapted to environmental variation.

Fecundity expresses fertility, which is one of the strategies developed by an organism to ensure its survival (Zouari, 2010). The absolute fecundity estimated for C. armatum in the PNIE is comparable to that recorded by Rotimi et al. (2012) for the same species, which ranges from 300,000 to 700,000 eggs. For Cardisoma crassum, Vasquez and Ramirez (2015) recorded between 173595 and 866857 eggs, with a mean of 501022 eggs in Mexico. The difference in fecundity between individuals of the species Cardisoma armatum, on the one hand, and between those of the species Cardisoma armatum and Cardisoma crassum, on the other, is explained by the size of the carapace and the body weight of the animal (Arimoro and Idoro, 2007; Lawal-Are, 2010; Olugbenga and Oloko, 2013). The larger the crab, the greater its mass and fecundity.

Analysis of the size of eggs on the abdomen of the crabs using the coefficient of variation showed that they have a heterogeneous structure. This suggests that *C. armatum* spawns fractionally.

Conclusion

The study of the reproductive parameters of *Cardisoma armatum* shows that the species reproduces twice a year, from June to October, with a spawning period from August to October, and from November to March, with a spawning period from January to March, and has a relatively high absolute fecundity. *Cardisoma armatum* therefore has quite interesting reproductive parameters and could be used as a farmed crab.

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