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Some aspects of reproductive biology and food ecology of lowlands Crabs *Liberonautes latidactylus* (De Man, 1903) - Côte d'Ivoire

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Abstract
Crabs, crustaceans of the order Decapoda, play an economic, dietary, bioecological, scientific and health role. Despite their important roles and their omnipresence in semi-terrestrial and aquatic environments, crabs receive less attention than other groups that are traditionally better studied, such as fish and shrimp. They have been largely neglected and few new studies have been conducted on them. Consequently, the present study focused on characterising the feeding ecology and reproductive biology of *Liberonautes latidactylus* in the shallows of the Adzopé department (Côte d'Ivoire). A total of 60 crab specimens, comprising 23 females and 37 males, were captured by hand and pitfall trapping from January to June 2024. The study of reproductive biology showed that the determined condition factor (K = 34%) indicates that environmental conditions are favourable for its development and that the recorded sex ratio 1.60 (M/F) favours males. Chi-square test revealed no significant difference sex ratio between seasons ($X^2 = 0.33$; $p = 0.56 > 0.05$). The feeding ecology of *Liberonautes latidactylus* revealed a species with a phytophagous diet with omnivorous tendencies. Macrophytes (IRA = 51.97%) ranked in the category of primary prey. Zooplankton (IRA = 18.49%) and Molluscs (IRA = 11.21%) are secondary prey. Worms (IRA = 8.16%) and nsects (IRA = 5.51%) were classified as accessory prey. Spearman's correlation test based on Relative Importance of Food (IRA) revealed that the food spectrum is not similar in males and females ($p = 0.77 > 0.05$). These results constitute preliminary data that could serve as a basis for further studies. They could be used as a reference in the sustainable management and captive breeding of this species.

Keywords: Fiddler crab, dietary profile, condition factor, sex ratio

1. Introduction
The hydrographic network of Côte d'Ivoire is home to a wide variety of crabs [1]. Crabs belong to the suborder Crustacea and are highly diverse in terms of their habitats, physiology and structure [2]. Although widely distributed on the seabed and along shorelines, crabs also inhabit freshwater and some species even venture onto land. Close relatives of lobsters and shrimps, they are distinguished by their evolution, which allows them to walk, run sideways, dig and swim with agility. Their bodies are protected by a chitinous carapace covered with a waxy coating. Crabs appear to be particularly evolved animals. With a relatively complex nervous system, they adapt effectively to changes in their environment, which helps them survive in hostile habitats [3]. There are more than 6,800 validated species and subspecies of crabs [4]. Crabs play an essential role in aquatic ecosystems, both economically and bioecologically, and occupy numerous trophic niches [5]. Some species are edible and widely exploited in aquatic ecosystems [6]. Like fish, crabs are among the most popular foods in Côte d'Ivoire [7] and represent an important food resource for Ivorians [8]. They are actively fished and mainly exploited for commercial purposes, thus constituting a source of income for local populations. They also oxygenate sediments by digging their burrows and therefore play a relatively important role in the anaerobic and aerobic mineralisation processes of sedimentary organic matter [9].

Certain crab species are used as indicators and/or biomarkers in assessing the quality of aquatic environments [10]. Given the importance of these crustaceans, several studies have been conducted on their biological and ecological aspects [2-11-12]. However, this research has focused solely on species of economic interest belonging to the genera *Callinectes* and *Cardisoma*. The mud crab *Liberonautes latidactylus* (De Man, 1903) is a species belonging to the family Potamonautidae and the subfamily Liberonautinae. This species is endemic to West Africa and is listed on the IUCN Red List in 2020 as locally threatened due to the degradation of waterways and mangroves, pollution, deforestation and health pressures [13]. *Liberonautes latidactylus* (De Man, 1903) is a large, robust riverine species, the most common and frequently caught in freshwater streams and rivers in savannah and forest areas. This species is sold in local markets and frequently consumed by local populations [14]. Furthermore, work carried out by [11] has shown that *Liberonautes latidactylus* is a species frequently observed in the terrestrial environment, which suggests that it probably moves easily between watersheds. However, no recent geographical studies have been undertaken on *Liberonautes latidactylus*, making it likely that new species exist in regions that have not yet been sampled. Despite the work carried out on crabs of the genus *Liberonautes*, no data exists on the diet and reproductive aspects of this species. The objective of this study is therefore to characterise the diet of *L. latidactylus* by analysing its variations according to sex and hydrological seasons, and also to address some aspects of its reproductive biology through sex ratio and condition factor in order to better understand the functional role of this species in Ivorian watercourses and to support conservation strategies.

2. Material and methods

2.1 Study area

The department of Adzopé is located in the south-east of Côte d'Ivoire in the Mé region between latitude 6.10° North and longitude 3.87° West. It covers an area of 3,724 km². Located 105 km from Abidjan, this agricultural area comprises six sub-prefectures, including Adzopé, Agou, Assikoi, Bécédi-Brignan, Yakassé-mé and Annépé. It is bordered to the north by the department of Akoupé, to the south by the departments of Abidjan and Alépé, to the east by the department of Yakassé-Attobrou, and to the west by the department of Agboville [15]. Three sites were selected for sampling: the village of N'Koupé and two low-lying areas in the town of Adzopé (Figure 1).

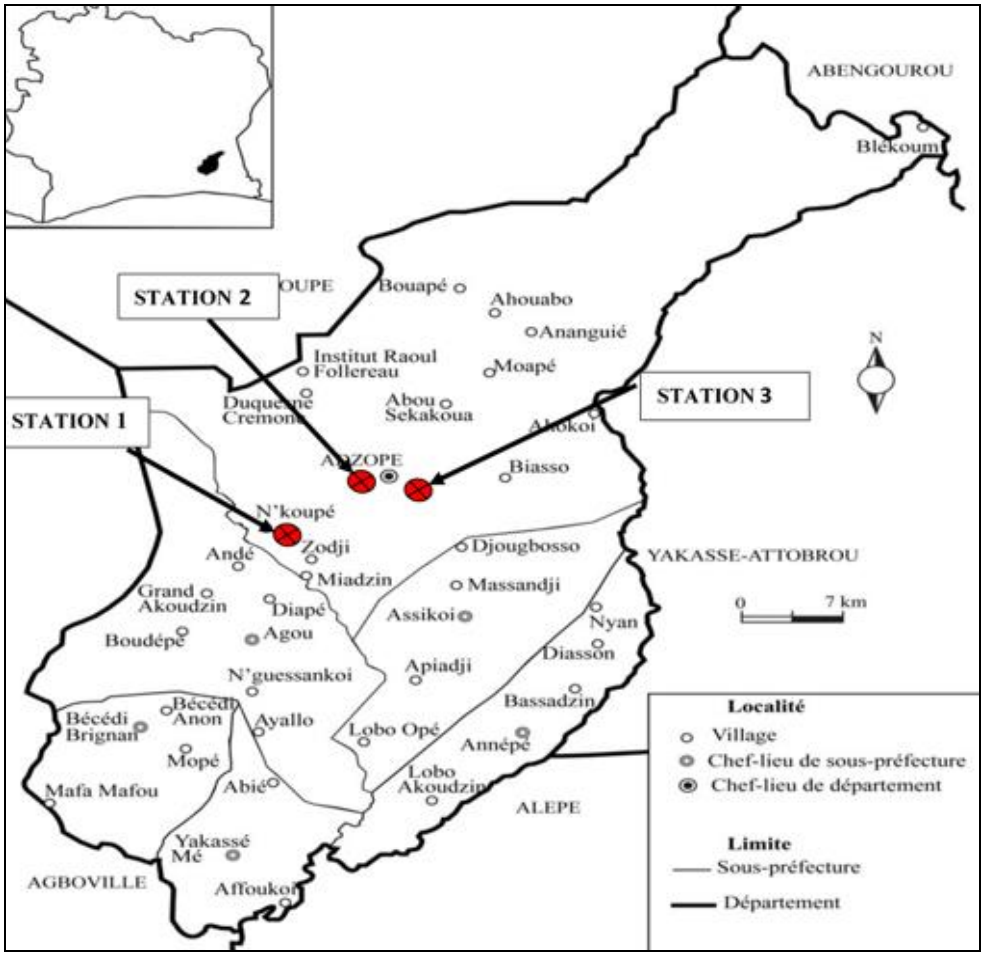

Fig 1: Location of sampling sites of *Liberonautes latidactylus* from January to June 2024

2.2. Sampling and laboratory examination

Manual sampling was carried out every month between January and June 2024 using a landing net and by digging crab burrows. At each sampling site, crabs were captured between 6 a.m. and 5 p.m. for daytime catches and between 6 p.m. and 10 p.m. for night-time catches. The crabs were identified according to [16-17]. The total carapace length (Lc) in mm of each individual was measured to the nearest 0.01 mm using a digital calliper (CD-15 APX; Mitutoyo, Kanagawa, Japan). The total weight (W) in g was recorded using a BP 310S top-loading balance (Sartorius, Göttingen, Germany) with an accuracy of 0.01 g. Each specimen was dissected in

the laboratory to remove the cardiac stomach and intestine. The cardiac stomachs were weighed and preserved in 5% formalin. The intestine was unrolled and measured in mm from the pylorus to the anus. The cardiac stomach was opened and its contents were observed using an SX25 binocular microscope (Bresser, Rhede, Germany). The prey were identified, counted and weighed using a Sartorius TE153S balance with an accuracy of 0.001 g. The prey were identified according to [18-19-20] for insects and crustaceans, [21] for molluscs, and [22-23] for fish.

2.3. Data analysis

1. **Condition factor study:** In this study, the condition factor (K) was calculated using the formula defined by [24]: $\mathbf{K} = \frac{\mathbf{M}}{\mathbf{LC^3}} \times \mathbf{100}$; M = crab mass (g), LC = carapace width (cm)
2. **Sex ratio:** In this study, the sex ratio (SR) was defined as the ratio of the number of males (M) to the number of females (F) at a given time [24]. Its formula is as follows: $\mathbf{SR} = \frac{M}{F}$; where M = number of males and F = number of females.
3. The vacuity coefficient (VC) was used to assess the intensity of the crabs' trophic activity. It is expressed as the percentage ratio between the number of empty stomachs (nev) and the total number of stomachs (nt) examined. $VC = \frac{\mathbf{n_{ev}}}{\mathbf{n_t}} \times \mathbf{100}$
4. The Relative Importance of Food (IRA) index developed by [25], combining three food indices (numerical percentage, weight percentage and corrected occurrence percentage), was used to assess the contribution of each category of items in the diet of *Liberonautes latidactylus*. $IRA = \frac{\%O_c + \%N + \%P}{\sum_1^5 (\%O_c + \%N + \%P)} \; x \; 100$; Oc = corrected occurrence percentage, %N = numerical percentage, %P = weight percentage and s = total number of food items. The different items were ranked in order of importance using the classification scale established by [25]. This classification is as follows: IRA ≥ 50%: main prey; 10% ≤ IRA < 50%: secondary prey; IRA < 10%: incidental or accidental prey.
5. The Chi-square test was used to compare the percentages of *L. latidactylus* sexes according to month and season. It was also used to compare the condition factor between the sexes of the crab studied and between seasons. Spearman's rank correlation coefficient test was used to compare the food spectrum of the species studied according to sex and season.

3. Results

3.1 Condition factor

The condition factor was determined on individual crabs with carapace widths ranging from 22.6 to 62.9 mm (Figure 2). The overall condition factor calculated is 34%. Depending on the hydrological season, the condition factor is higher in males and females during the dry season, with values of 38% and 32% respectively. In the rainy season, it varies between 34% for males and 33% for females. The condition factor for both sexes combined is higher in the dry season (35%) than in the rainy season (34%). Statistical analysis reveals no significant difference in the condition factor between seasons ($p = 1 > 0.05$; Chi-square test).

Taking sex into account, the mean value of the condition factor K is 0.35 ± 0.1 in males compared to 0.32 ± 0.09 in females. Statistical analysis reveals no difference in body weight between the sexes of *Liberonautes latidactylus* ($p = 1 > 0.05$; Chi-square test).

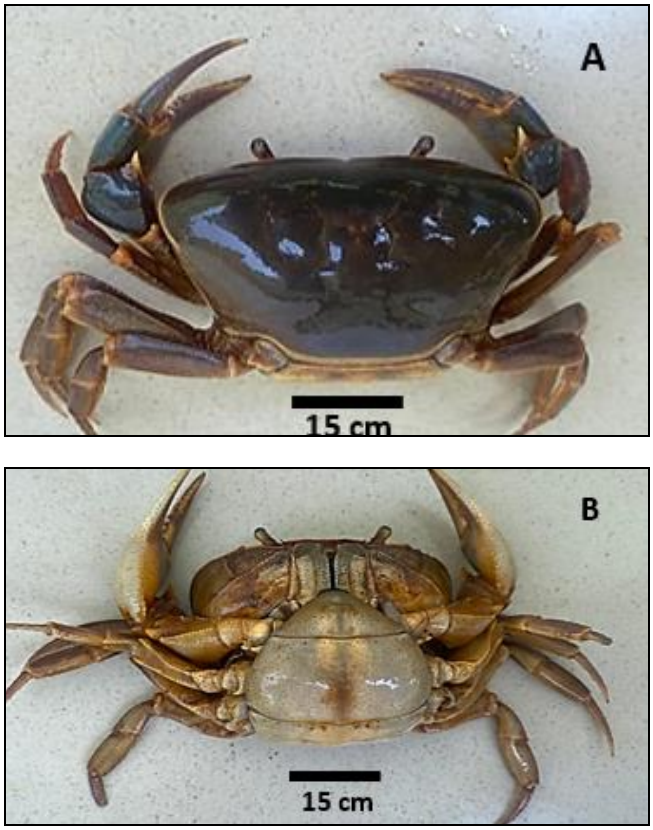

Fig 2: *Liberonautes latidactylus* sampled in the shallows areas from January to June 2024 (A: Ventral side; B: Dorsal side)

3.2 Sex ratio

A total of 60 individuals of *L. latidactylus* were sampled, including 37 males and 23 females. The calculated masculinity rate is 61.67% and the femininity rate is 38.33%. The overall sex ratio was 1.60 (M/F), favouring males. During the dry season, 20 crabs were captured, including 7 females and 13 males, giving a masculinity rate of 65% and a femininity rate of 35%. The sex ratio during the dry season is 1.86. During the rainy season, 40 crabs were captured, including 16 females and 24 males, giving a masculinity rate of 60% and a femininity rate of 30%. The sex ratio is 1.5. The Chi-square test revealed no significant difference between seasons ($X^2 = 0.33$; $p = 0.56 > 0.05$).

3.3 Vacuity coefficient

A total of 60 *Liberonautes latidactylus* crabs were selected for stomach content analysis. Of this total, 3 crabs had empty stomachs, giving an overall vacuity coefficient of 5%. Analysis of stomach filling rates showed variation in this parameter depending on the season in which the crabs were caught. In the rainy season, 1 in 40 crabs had empty stomachs, giving a vacuity coefficient of 2.5%. In the dry season, 2 in 20 crabs had empty stomachs, giving a vacuity coefficient of 10%.

3.4 General profile of diet

Qualitative analysis of stomach contents recorded in stomachs and the cardiac tube showed that the dietary spectrum of *L. latidactylus* consists of animal and plant prey and sediments (Table 1). The animal fraction consists of insects, molluscs,

worms and plankton. The insects are represented by remains including wings, legs and integuments. The molluscs consist of snail shells. The worms recorded are annelids. Plankton is represented by rotifers and cladocerans. The plant fraction consists mainly of plant debris. Sediments are represented by mud and sand grains. The highest percentage by weight was observed in macrophytes, at 87.03%. Analysis of the numerical proportions of prey shows that zooplankton (N = 30.91%) is the most abundant prey in the stomach of *L. latidactylus*. It is followed by macrophytes (N = 30.3%), worms (N = 13.94%), molluscs (N = 12.12%), sediments (N = 6.67%) and insects (N = 6.06%). The classification of prey based on Relative Importance of Food (IRA) allowed Macrophytes (IRA = 51.97%) to be ranked in the category of primary prey. Zooplankton (IRA = 18.49%) and Molluscs (IRA = 11.21%) are secondary prey. Worms (IRA = 8.16%) and insects (IRA = 5.51%) were classified as accessory prey in the food spectrum of *L. latidactylus.*

Table 1: General composition of the diet of *Liberonautes latidactylus* sampled in the shallows of the Adzopé department (Côte d'Ivoire) from January to June 2024

Items categories	%Oc	%P	%N	%IRA
Macrophytes				
Plant debris	38.6	87.03	30.3	51.97
Insects	8.77	1.71	6.06	5.51
Molluscs				
Snail shells	12.28	9.21	12.12	11.21
Worms				
Annelids	10.53	0	13.94	8.16
Zooplankton				
Cladocera	7.02	0	15.76	7.59
Rotifera	17.54	0	15.15	10.9
Sediment				
Sand grains	5.26	2.05	6.67	4.66
Macrophytes	38.6	87.03	30.3	51.97
Insects	8.77	1.71	6.06	5.51
Molluscs	12.28	9.21	12.12	11.21
Worms	10.53	0	13.94	8.16
Zooplankton	24.56	0	30.91	18.49
Sediment	5.26	2.05	6.67	4.66

% Oc = Percentage of occurrence; % P = Percentage by weight; % *n* = Percentage by number; IRA = Relative Importance of Food

3.5 Variation in diet according to sex

The analysis of the food spectrum of *L. latidactylus* according to sex was based on 36 males and 21 females with full stomachs. These results show that both males and females feed on macrophytes, insects, molluscs, zooplankton and worms (Table 2). The highest percentage by weight is observed in macrophytes for both males and females, with respective values of 87.20% and 86.76%.

In males, zooplankton is the most important with a numerical percentage of 37.26%, followed by macrophytes (*n* = 27.31%), worms (*n* = 12.72%) and molluscs (*n* = 10.9%). Based on occurrence, the most regularly consumed prey are macrophytes (Oc = 38.94%). In females, macrophytes are the most abundant with N = 36.37%. They are followed by zooplankton (*n* = 18.18%), worms (N = 16.36%) and molluscs (*n* = 14.55%). The most regularly consumed prey are macrophytes (Oc = 33.43%), molluscs (Oc = 23.81%) and worms (Oc = 19.05%).

The classification of prey based on Relative Importance of Food (IRA) indicates that macrophytes, with values of 51.27% in males and 52.15% in females, are the main prey. Plankton (IRA = 17.6%), molluscs (IRA = 11.94%) and worms (IRA = 10.73%) represent secondary prey for males. For females, only molluscs (IRA = 16.09%) and worms (RFI = 11.81%) are secondary prey. Other prey categories with an IRA of less than 10%, consisting of insects (both sexes combined) and zooplankton (in females), are consumed incidentally.

Spearman's correlation test based on Relative Importance of Food (IRA) revealed that the food spectrum is not similar in males and females ($p = 0.77 > 0.05$).

Table 2: Diet composition of *Liberonautes latidactylus* according to sex sampled in the shallows of the Adzopé department (Côte d'Ivoire) from January to June 2024

	Males (n = 36)				Females (n = 21)			
Items categories	**%Oc**	**%P**	**%N**	**%IRA**	**%Oc**	**%P**	**%N**	**%IRA**
Macrophytes								
Plant debris	38.94	87.20	27.31	51.27	33.43	86.76	36.37	52.15
Insects	5.56	1.74	5.45	4.25	9.52	1.65	7.27	6.15
Molluscs								
Snail shell	16.67	8.71	10.9	12.1	23.81	9.92	14.55	16.09
Worms								
Annelids	19.44	0.01	12.72	10.73	19.05	0.01	16.36	11.81
Zooplankton								
Cladocera	5.56	0.01	14.54	6.7	9.52	0.01	18.18	9.24
Rotifer	8.33	0.01	22.72	10.36	0	0	0	0
Sediment								
Sand grains	5.56	2.32	6.36	4.75	4.76	1.65	7.27	4.56
Macrophytes	38.94	87.20	27.31	51.27	33.43	86.76	36.37	52.15
Insects	5.56	1.74	5.45	4.25	9.52	1.65	7.27	6.15

Molluscs	16.21	8.71	10.9	11.94	23.81	9.92	14.55	16.09
Vers	19.44	0.01	12.72	10.73	19.05	0.01	16.36	11.81
Zooplankton	13.89	0.02	37.26	17.06	9.52	0.01	18.18	9.24
Sediment	5.56	2.32	6.36	4.75	4.67	1.65	7.27	4.56

% Oc = Percentage of occurrence; % *p* = Percentage by weight; % *n* = Percentage by number; IRA = Relative Importance of Food.

Variation in diet according to seasons

The composition of the diet of *L. latidactylus* according to the hydrological seasons revealed consumption of macrophytes, insects, worms, zooplankton, and molluscs in both the dry and rainy seasons. Macrophytes had the highest percentage by weight in both the rainy season (*p* = 87.62%) and the dry season (*p* = 85.83%) (Figure 3). Analysis of numerical proportions showed that during the rainy season, the prey most consumed by *L. latidactylus* are zooplankton (*n* = 34.29%) and macrophytes (*n* = 30.48%). They are followed by worms (*n* = 14.28%) and molluscs (N = 12.28%). Insects are poorly represented, with a numerical importance of N = 5.71%. In terms of occurrence, macrophytes (Oc = 30.77%) and zooplankton (OCc = 28.02%) are the most frequent prey in the stomach of *L. latidactylus.* During the dry season (SS), *L. latidactylus* consumes 85.83% macrophytes, followed by 8.08% molluscs, 2.02% insects, 0.02% zooplankton and 0.01% worms. However, zooplankton (%Oc = 27.78%) remains the most frequently consumed prey during this period.

The classification of prey based on Relative Importance of Food (IRA) showed that no prey can be considered the main prey due to their IRA being less than 50%. Macrophytes (IRA = 49.62%), zooplankton (IRA = 20.83%) and molluscs (IRA = 11.66%) are secondary prey consumed during the rainy and dry seasons. In the rainy season, insects (IRA = 4.98%) and worms (IRA = 9.89%) are considered incidental prey. In the dry season, only insects (IRA = 9.89%) are incidental prey. Spearman's rank correlation test based on Relative Importance of Food showed that the diet of L. *latidactylus* is similar for both seasons (p = 0.004 < 0.05).

During the rainy season, males (IRA = 48.69%) and females (IRA = 48.97%) mainly consume macrophytes. During the dry season, males and females with respective values of IRA = 47.49% and IRA = 43.04% mainly feed on macrophytes (Figure 4).

Plankton (IRA = 11.12%), molluscs (IRA = 10.55%) and worms (IRA = 12.51%) are secondary prey consumed by females during the dry season. Insects (IRA: males = 5.29% and females = 4.47% in the rainy season and IRA: males = 5.11% and females = 9.44% in the dry season) are incidental prey. Spearman's rank correlation test showed that the food spectrum of L. *latidactylus* is not similar for both seasons in females (*p* = 0.25 > 0.05) but similar in males (*p* = 0.04 < 0.05).

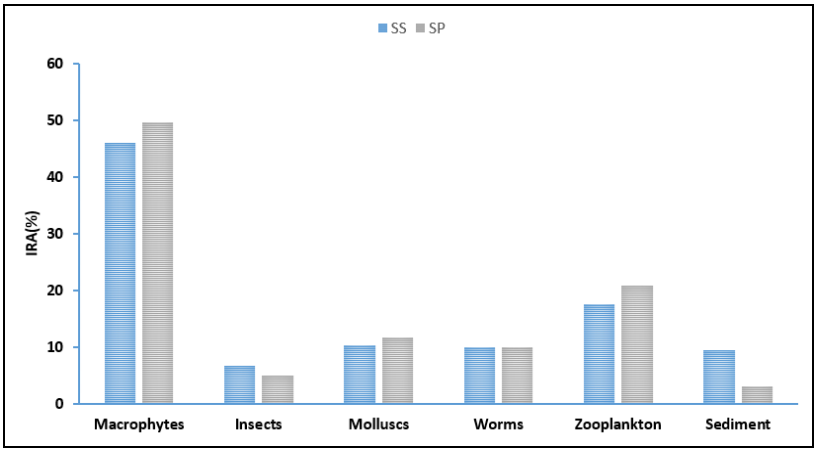

Fig 3: Variation in food spectrum according to season in *Liberonautes latidactylus* sampled in the shallows of the Adzopé department (Côte d'Ivoire) from January to June 2024. SS = Dry season and SP = rainy season

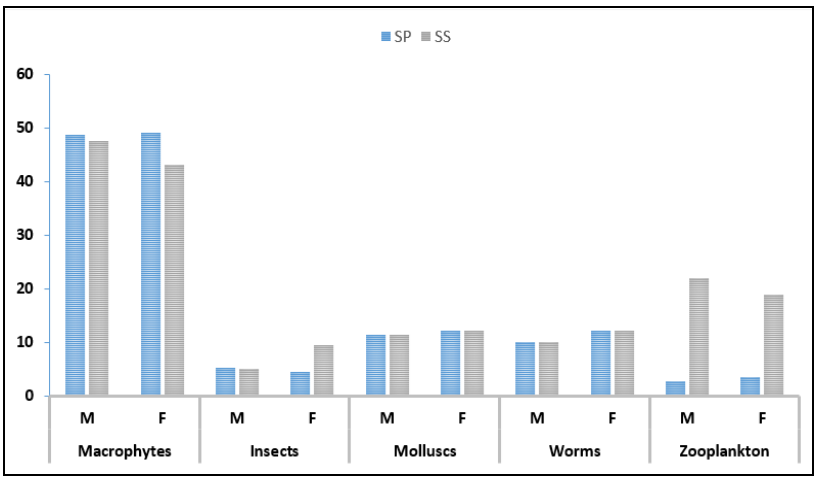

Fig 4: Variation in food spectrum according to sex and season in *Liberonautes latidactylus* sampled in the shallows of the Adzopé department (Côte d'Ivoire) from January to June 2024

Discussion

The condition coefficient K allows us to estimate seasonal changes in body weight under the influence of external and internal factors [26]. The condition factor for L. *latidactylus* of both sexes combined is 0.34. This high value for body condition shows that L. *latidactylus is* well adapted to the environmental and biological conditions of its habitat. Males were more overweight than females, meaning that males grow faster than females. These results are consistent with the work of [27], who reported higher condition factors in males of *Portunus validus* off the coast of Lagos, Nigeria. In contrast, [28] reported higher condition factor values in females of *Callinectes amnicola* on the Warri River in Nigeria. These observations show that the condition factor value varies from one species to another and changes according to morphology, sex, age, and reproductive status associated with variations in gonadal maturity stages [29]. Crabs caught in the shallows of Adzopé show a dominance of *L. latidactylus* males over females. The overall sex ratio calculated is 1.60. Similar results were observed by [30] in Nigeria in *Cardiosoma armatum*, with a sex ratio of 1.12. On the other hand, a sex ratio favouring females was recorded by [31] in Nigeria in C. *armatum*, with a value of 1.18. These observations show that the sex ratio can change within the same species. This variation could be explained by genetic, environmental [32], physiological and hormonal, ecological factors, as well as selective pressures and evolutionary strategies of species [33-34]. The general analysis of the food spectrum of *L. latidactylus* shows an overall vacancy rate of 5%. The temporal study of the vacancy index reveals that *L. latidactylus* exhibits a seasonal pattern of feeding activity in shallow waters. The percentage of vacuity is relatively higher in the dry season than in the rainy season, with respective values of 10% and 2.5%. These vacuity percentages are high compared to those obtained by [12] in the Ehotilé Islands National Park, where he obtained a vacuity percentage of 1.97% for *C. armatum.* In the present study, the high vacuity percentages could b ly explained by low trophic activity of the species, the time of collection, the biological phase (moulting or reproduction) and/or the time elapsed before freezing. Indeed, *L. latidactylus* is predominantly nocturnal, so daytime catches often show a very high vacuity rate [35-36]. The food spectrum of *L. latidactylus* includes a variety of prey consisting of macrophytes, insects, molluscs, plankton and worms. Grains of sand and mud have also been observed in stomach contents. Our results are comparable to those of [30], who observed *C. armatum* in rivers in Nigeria. The variety of prey obtained in this study showed that *L. latidactylus* has a phytophagous diet with omnivorous tendencies. Similar observations were made by [37] in *C. armatum* and *C. guanhumi.* The classification of prey based on Relative Importance of Food (IRA) showed that the main prey are macrophytes with 51.97%, followed by plankton and molluscs, which are secondary prey with respective IRA of 18.49% and 11.21% in *L. latidactylus.* The high consumption of macrophytes is thought to be due to the availability and/or selectivity of this food category in the study area. The results of this study are comparable to those obtained by [38-39], which showed a preference for plant debris in the mangrove crab *Sesarma huzardi*. The sediment consisting of mud and sand grains observed in the stomachs shows that *L. latidactylus* feeds in the mud. Indeed, sand grains could play a role in grinding the food consumed by this species. Analyses of the food spectrum according to sex showed that plankton prey was numerically most important in males with 37.26%, while macrophyte prey was numerically most abundant in females with 36.37%. However, macrophytes are the main prey consumed by both sexes. 's Spearman's rank correlation test based on Relative Importance of Food (IRA) did not reveal any similarity in the food spectrum between the sexes ($p = 0.77 > 0.05$). The lack of similarity in the food spectrum between the sexes could be explained by prey selectivity linked to a strong ability to search for food, particularly plankton, which is consumed in abundance by males [37]. The composition of the food spectrum of *L. latidactylus* according to the seasons revealed that the prey most abundantly consumed by *L. latidactylus* in the rainy season is plankton, with 34.29%, and in the dry season, macrophytes, with 30%. The abundance of plankton in the rainy season can be explained by the accessibility and availability of these prey [37]. Considering the seasonal variation in the food spectrum according to sex, it appears that males and females mainly consume macrophytes (IRA = 48.69%; IRA = 48.97% respectively) during the rainy season and dry season (IRA = 47.49% and IRA = 43.04% respectively), with a significant difference between the two seasons in females ($p = 0.25 > 0.05$) but similar among males ($p = 0.04 < 0.05$). These results could be explained by seasonal availability and selectivity of prey by females. Indeed, a decrease in the consumption of primary prey from one season to another is compensated by secondary prey. These observations are similar to those of [40] on *Trichodactylus borellianus* (Trichodactylidae) from South America, which showed that the diets of juvenile, subadult and adult crabs differ in their capacities and preferences.

5. Conclusion

Liberonautes latidactylus is a species of freshwater mud crab. Studies of its diet have revealed a highly diverse diet consisting of macrophytes, insects, molluscs, plankton and worms. Macrophytes are the most abundant and mainly consumed food source. L. *latidactylus* is therefore a species with a phytophagous diet and omnivorous tendencies. Further research providing more details on the feeding behaviour of the species, taking into account the differences between the microhabitat s of juveniles and adults and how these habitats influence their feeding habits, will help to define a policy for the conservation and preservation of the species. This information is particularly necessary given that the species is facing significant population disruption due to the excessive exploitation of shallow waters with the development of rice cultivation, market gardening, the misuse of plant protection products and the destruction of its habitat.

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