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Zooplanktonic dietary preferences of *Ethmalosa fimbriata* (Clupeidae) and *Eucinostomus melanopterus* (Gerreidae) in Lake Nokoué in Benin

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ABSTRACT

The aim of the present study was to examine the zooplanktonic prey preferences of two important artisanal fish species, *Ethmalosa fimbriata* (Clupeidae) and the *Eucinostomus melanopterus* (Gerreidae) in Lake Nokoué. A monthly sampling was conducted from September 2021 to January 2022, and the stomach contents of the sampled fish were analyzed using presence-absence and numeric abundance methods. The results revealed that both species primarily consume three main zooplankton taxa: copepods, rotifers, and cladocerans. *Ethmalosa fimbriata*, especially individuals subadults (9-13 cm) consistently favored Copepods, with an occurrence frequency of 70.45%, regardless of its size. In contrast, *Eucinostomus melanopterus*, especially adult individuals (13-15 cm), exhibited a strong preference for Cladocerans, with an occurrence frequency of 50.54%. The monthly variation in the Average Nutrition Index (ANI) for *E. fimbriata* showed higher feeding intensity during November (5.9 ± 5.0) and lower in January (1.92 ± 1.2), with a significant difference *(P-value<0.05)*. Principal Component Analysis indicated no significant zooplankton prey preference based on size among juvenile stages of *E. fimbriata*. However, adult *E. melanopterus* demonstrated a discernible selectivity towards Cladocerans. These findings, with significant size-dependent variations *(P-value<0.001)*, provide valuable insights into the dietary preferences and ecological roles of these two fish species in Lake Nokoué.

Keywords: Copepods, Cladocerans, Rotifers, White Flag, African Ethmalosa, stomach contents.

INTRODUCTION

Aquatic ecosystems shelter diverse organisms such as plankton and fish which interact each other and with their environment (Adandedjan et al., 2017). In West African coastal water bodies, such as estuaries, lagoons and the Gulf of Guinea, *Ethmalosa fimbriata* (Clupeidae) commonly called African Ethmalosa, West African shad or Bonga shad is one of abundant pelagic fish species that

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plays a vital role in insuring the nutritional needs of local populations (Ajah and Asuquo, 2017; Baldé et al., 2018; Palomares et al., 2020; N'Dri et al., 2023). *Eucinostomus melanopterus* (Gerreidae), also known as the white flag or flag-fin mojarra, is another important fish species commonly found in many West African coastal water bodies (Udoh and Ekpo, 2017; Assou et al., 2018; Eugenia et al., 2019; Samake et al, 2022). In Lake Nokoué, these two species are abundant in artisanal fishing catches, representing approximately half of the total ichthyofauna abundance, with 40% attributed to *Ethmalosa fimbriata* and 10% to *Eucinostomus melanopterus* (Djihouessi et al., 2019). To ensure a sustainable management of these two fishes' stock in the lake in relation with the lake environmental conditions changes in the one hand and to promote their sustainable farming in another hand, accurate information on their feeding behavior is needed. Indeed, according to Amundsen and Sánchez-Hernández (2019), knowledge about fish nutrition in natural environment is essential for comprehension of their biology, trophic ecology, relationship between fish populations and ecosystems functioning.

Several recent studies were focused on the food diet of *Ethmalosa fimbriata* (Clupeidae) and *Eucinostomus melanopterus* (Gerreidae) in some West African countries such as Ghana, Mauritania, Nigeria and Senegal (Gushchin, 2013; Ekpo et al., 2014; Ndour et al., 2017; Udoh and Ekpo, 2017; Baidoo, 2018). In Benin, they are part of the zooplanktivorous fish community in Lake Nokoué (Gnohossou et al., 2013). Zooplankton, are known to ensure transfer of the primary production (from phytoplankton) to superior food levels including fish and particularly fish juvenile (Karpowicz et al., 2020). Knowledge of zooplankton prey can then be useful in the development of aquaculture programs for these fishes and as a foundation for research on live prey feeding for these species. But the main zooplankton taxonomic groups, prey, of those species are unknown in Lake Nokoué that constitutes the most important and productive (65 to 70% of fish resources) continental water plan in Benin (Gnohossou, 2006; Degila et al., 2020; Chaigneau et al., 2023). Gnohossou et al. (2013) studied the general food diet of these two species but did not investigate their plankton diet from a taxonomic perspective. Therefore, this study was carried out to examine specifically the zooplankton consumed by *Ethmalosa fimbriata* and *Eucinostomus melanopterus*, providing a detailed taxonomic analysis of their planktonic prey in Lake Nokoué.

MATERIALS AND METHODS

Study area

Ethmalosa fimbriata and *Eucinostomus melanopterus* were collected at 12 stations from Lake Nokoué (Figure 1). This lake is situated between parallels 6 ° 20 ' N and 6 ° 30 ' N and meridians 2 ° 20 ' E and 2 ° 35 ' E, making it the largest lake in Benin, covering approximately 150 km² during the dry season (Adandedjan et al., 2017; Chaigneau et al., 2023). Lake Nokoué is bounded to the West by Abomey-Calavi plateau, to the East by Porto-Novo lagoon, to the North by the flooded plains of Ouémé and Sô Rivers, and to the South by Cotonou town (Degila et al., 2020). It is connected to the Atlantic Ocean and Porto-Novo lagoon through Cotonou and Totché channels, respectively (Sachi et al., 2016; Adandedjan et al., 2017; Chaigneau et al., 2023).

Lake Nokoué experiences four distinct seasons in its hydrology : low floods from May to June, corresponding to the high rainy season in southern Benin; high floods from September to November, corresponding to the low rainy season and water coming from Ouémé River and river Sô; low dry period from December to March and low dry season from mid-July to mid-September (Adandedjan et al., 2017). Flood duration varies based on the opening or closing of Cotonou channel, influenced by marine sand accumulation and rainfall intensity (Adandedjan et al., 2017). This alternation enables important variations of physicochemical parameters such as salinity, temperature, turbidity, dissolved oxygen and Chlorophyll-a into the lake (Chaigneau et al., 2023).

The zooplankton community in Lake Nokoué is primarily dominated by copepods, rotifers, and cladocerans (Adandedjan et al., 2017; Chaigneau et al., 2023). Chaigneau et al. (2023) inventoried 109 zooplankton species in the lake, including 81 rotifer taxa, 20 copepod taxa, five cladoceran taxa, and three taxa from other zooplankton groups (Eumalacostraca, Mollusca, and Ostracoda). Notably, rotifers from the families Brachionidae, such as *Brachionus plicatilis*, and Synchaetidae are the most prevalent in the lake (Adandedjan et al., 2017; Chaigneau et al., 2023). Among copepods, species from the orders Cyclopoida and Calanoida are the most commonly found and predominant (Adandedjan et al., 2017; Chaigneau et al., 2023).

Fish sampling and length measurements

Fish sampling was conducted monthly, spanning from September 2021 to January 2022, corresponding to low rainy season and low dry season. The collection process involved the collaboration with local fishermen, who assisted in both day and night fishing activities using nets known as "Medopokonou" (Gnohossou et al., 2013). These nets characteristics are: length 20 meters; mesh size 10 mm. Fish were identified in the field, using keys developed by Lévêque et al. (1990) and Lévêque and Paugy (2006) but in case of uncertainty or doubt regarding the identification, specimens were preserved fresh in icebox for detailed examination and identification in the laboratory. In the laboratory, the fish were subjected to a detailed morphological examination, including the observation of external characteristics and precise biometric measurements. If necessary, an examination of internal characteristics was conducted.

After identification, the specimens underwent a series of measurements, including weight, standard length (SL), fork length (FL), and total length (TL).

Stomach content analysis methods

The analysis of stomach contents was conducted following a systematic procedure. Each specimen was dissected on the sampling day, then the stomachs were extracted and weighed. Stomach contents were discharged from the dissected stomach, into a Petri dish for observation. Zooplanktonic preys were recognized using an OPTIKA B-290TB Digital Microscope (Optika Microscope, Milano, Italy) and identified to the lowest taxonomic level possible using identification keys provided by Glime (2017a, 2017b). The zooplankton identified were then counted.

The stomachs that were not analyzed on the sampling day were immediately frozen in the freezer for future analysis. When ready for analysis, the frozen stomachs were thawed, rinsed thoroughly with water, and then dried using tissue paper before dissection after removal from the freezer.

200 individuals of *E. fimbriata* weighing between 1.1 and 28.4 g and 212 individuals of *Eucinostomus melanopterus* weighing between 1.8 and 29.4 g, both with a size ranging from 5 to 15 cm, were sampled. Several indexes were used in the stomach content analysis such as:

Average Nutrition Index (ANI): ANI measures food intensity in fish. Variations of ANI provides insight into food cycle determination and prey quantity fluctuations over time. It is calculated by the following formula:

$$\text{ANI} = \frac{\text{Weight of stomach content}}{\text{Full weight of fish}} \times 100$$

For the zooplankton prey selectivity assessment, the presence-absence method and the numerical method as recommended by Amundsen and Sánchez-Hernández (2019) were used.

Occurrence frequency (OF%): This index represents the number of stomachs containing at least one individual of the prey (n) divided by the non-empty (N) stomachs total number.

$$\text{OF\%} = \frac{n}{N} \times 100 \text{ (Hureau, 1970)}$$

Based on this index, preys were classified in three categories by the following correspondences: preferred preys (OF% ≥

50%), secondary preys (10% <OF%< 50%) and occasional preys (OF% ≤ 10%) (El Achi et al., 2021).

Numeric abundance (N%): N% provides information on the feeding behavior of fish and is the ratio between the number of individuals of a given prey species (Np) and the total number of prey items (Ntp).

$$N\% = \frac{Np}{Ntp} \times 100 \text{ (Hyslop, 1980)}.$$

Statistical analysis

In order to test the normality of the data distribution the Shapiro-Wilk's test was used. Comparisons of Average Nutrition Index between sampling months/size groups were performed using the Analysis of Variance (One-way ANOVA). Since data sets lacked normality of distribution, the non-parametric Kruskal-Wallis test was used. Significance level (α) for all conducted tests was at 5%. In order to assess the relative similarity of food preferences among the different size classes of both species, Principal Components Analysis (PCA) was conducted as graphic method via the package FactorMineR (Lê et al., 2008) through RStudio software version 4.3.2 (R Core Team, 2023).

The analysis was based on data provided in a table containing occurrence frequencies of different groups of zooplankton prey (in column) according to size classes (in rows). *E. melanopterus* individuals were allocated to three different size classes (juvenile: less than 9 cm, sub-adult: 9-13 cm and adult: more than 13 cm), following the classification proposed by Ramos et al. (2014).

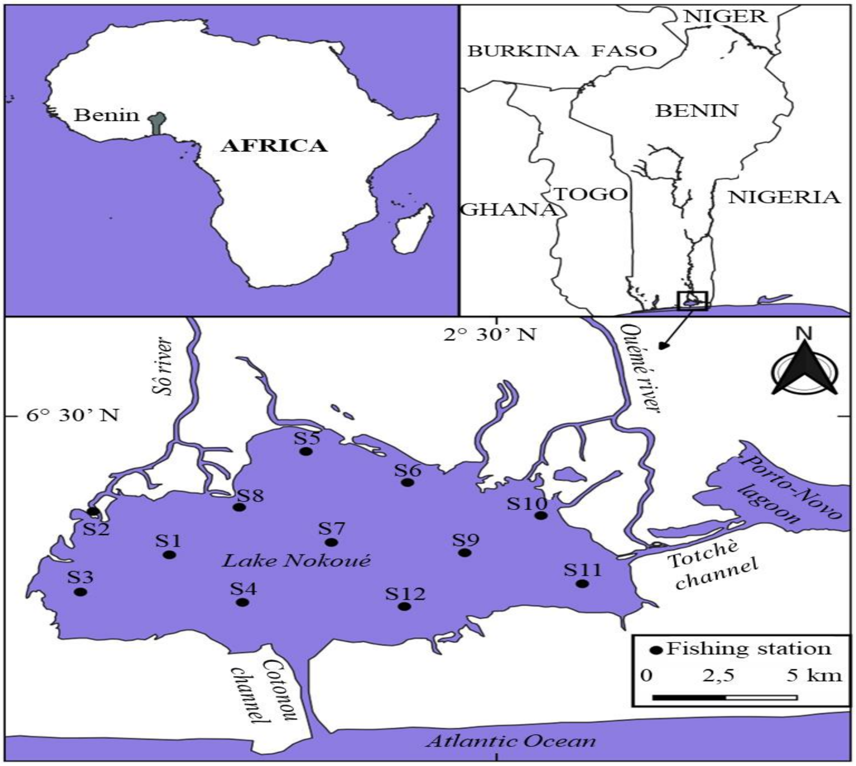

Figure 1: Map of Lake Nokoué with sampling sites in Benin.

RESULTS

Zooplanktonic prey and nutritional analysis of *Ethmalosa fimbriata*

Among the 200 individuals of *Ethmalosa fimbriata* examined, 176 individuals conained 413 zooplankton individuals in their stomachs. Data on the individual number of zooplanktonic preys consumed by *Ethmalosa fimbriata,* occurrence frequencies (FO) relative to these preys and their numeric abundance are presented in Table 1. The results show that the copepods occurrence frequency (FO = 70.45%) is greater than 50% indicating that copepods are the preferred zooplanktonic prey for *Ethmalosa fimbriata* in lake Nokoué. They were followed by rotifers (FO = 31.81%), cladocerans (FO = 27.84%), and undifferentiated zooplankton (FO = 11.93%) which were considered secondary zooplanktonic preys. This trend was confirmed by the numeric abundance data (59.81%), showing that copepods represented the largest quantity of consumed prey.

The monthly variation of *Ethmalosa fimbriata* average nutrition index is illustrated in Figure 2.

A peak was observed in November with a value of 5.9 ± 5.0, while January exhibited the lowest consumption, with a value of 1.92 ± 1.2. Both months show respectively high and low food intensity in *Ethmalosa fimbriata* during the study period with a significant difference *(P<0.05)*. Additionally, examination of the variations related to size classes showed that ANI decreased as fish size increased (Figure 3). For instance, ANI was 6.06 ± 5.72 for fish sizes ranging from 5-7 cm, but it decreased to 1.2 ± 0.62 for fish sizes between 13 and 15 cm, with a highly significant difference *(P<0.001)*, showing a size-dependent variation in nutrition.

Assessment of the similarity of occurrence frequencies of zooplanktonic prey among the different size classes of *Ethmalosa fimbriata,* using Principal Component Analysis, showed that the diet of *Ethmalosa fimbriata* individuals was not strongly associated with their size. The analysis grouped the different size classes into three main groups. The first group consisted of *E. fimbriata* individuals, ranging from 9 to 13 cm, who fed mainly on copepods. The second group was constituted of individuals whose size ranged from 7-9 cm and 13-15 cm that prefer rotifers and cladocerans. *E. fimbriata* small size individuals (5-7 cm), constituted the third group and had no clear preference (Figure 4).

Zooplankton consumption patterns of *Eucinostomus melanopterus*

Among the 212 individuals of *Eucinostomus melanopterus* examined, 188 individuals contained a total of 489 zooplankton prey items in their stomachs. Number of prey individual consumed by *Eucinostomus melanopterus*, their occurrence frequencies and numeric abundance are shown in Table 2. From the Table 2, Cladocerans had an occurrence frequency (OF= 50.54%) slightly higher than 50%, showing that they were preferred preys for *Eucinostomus melanopterus* in Lake Nokoué. They were followed by copepods (OF= 39.01%), rotifers (OF=23.07%) and non-identified zooplankton (OF=8.79%) with occurrence frequencies less than 10 making them occasional preys.

Assessment of zooplanktonic prey occurrence frequencies similitude level between different size classes of *Eucinostomus melanopterus* through the principal components analysis showed that feeding of *E. melanopterus* did not clearly depend on fish individual size (Figure 5). There was not a prey group only consumed by a unique individual size class. Many size classes fed on the same prey group.

Both graphics enabled to gather different classes in two big groups. The first group was made of juveniles and sub-adults (individuals ranging from 5-13 cm) that essentially fed on rotifers and copepods. Group of *E. melanopterus* adults whose individuals size vary from 13-15 cm, preferred cladocerans.

Table 1: Zooplankton preys of *Ethmalosa fimbriata* (Clupeidae) from the Lake Nokoué from September 2021 to January 2022.

Preys	**Ni**	**N%**	**OF%**
Copepods	247	59.81	70.45
Rotifers	57	13.80	31.81
Cladocerans	75	18.16	27.84
unidentified zooplankton	34	8.23	11.93
Total	413	100	

Ni = number of individuals of the prey, OF = occurrence frequency, N = numeric abundance.

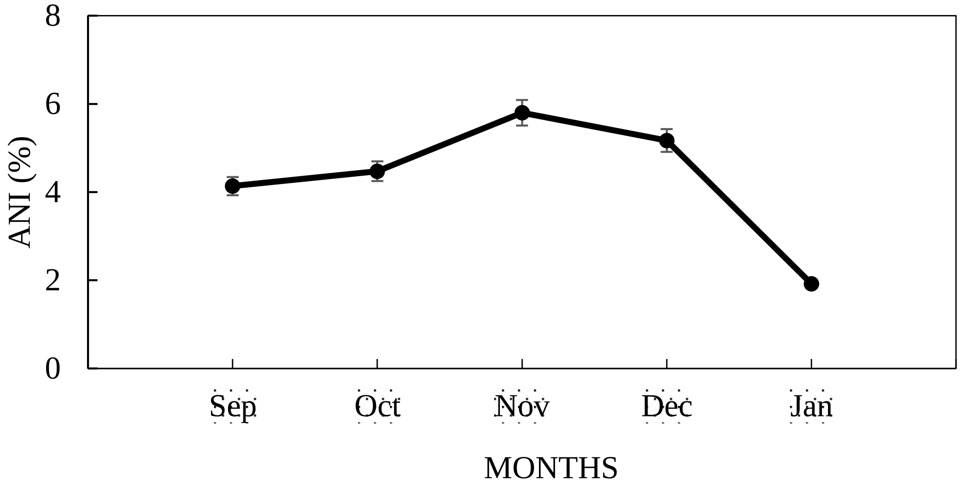

Figure 2: Monthly Average Nutrition Index variation in *Ethmalosa fimbriata.*

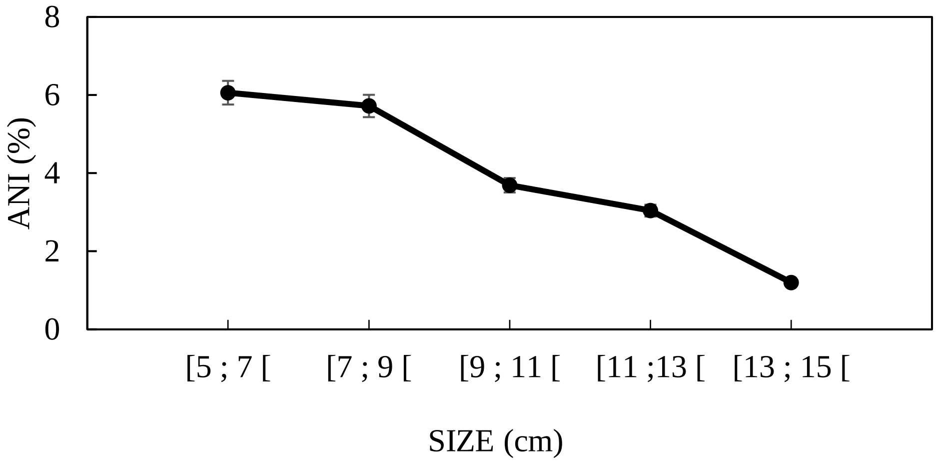

Figure 3: Variation of *Ethmalosa fimbriata* average nutrition index in relation to size.

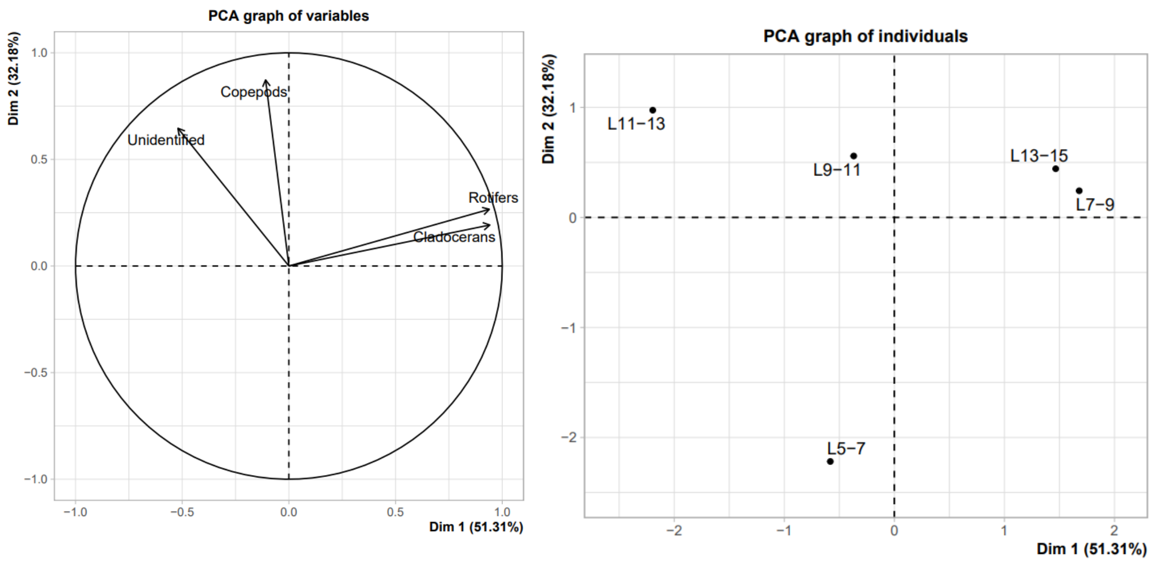

Figure 4: PCA variables graph and PCA individuals graph showing food preference similarities among size classes of *Ethmalosa fimbriata*.

Table 2: Zooplankton preys of *Eucinostomus melanopterus* (Gerreidae) from the Lake Nokoué from September 2021 to January 2022.

Preys	Ni	N%	OF%
Copepods	160	32.72	39.01
Rotifers	81	16.56	23.07
Cladocerans	226	46.22	50.54
Unidentified zooplankton	22	4.50	8.79
Total	489	100	

Ni = number of individuals of the prey, OF = occurrence frequency, N = numeric abundance.

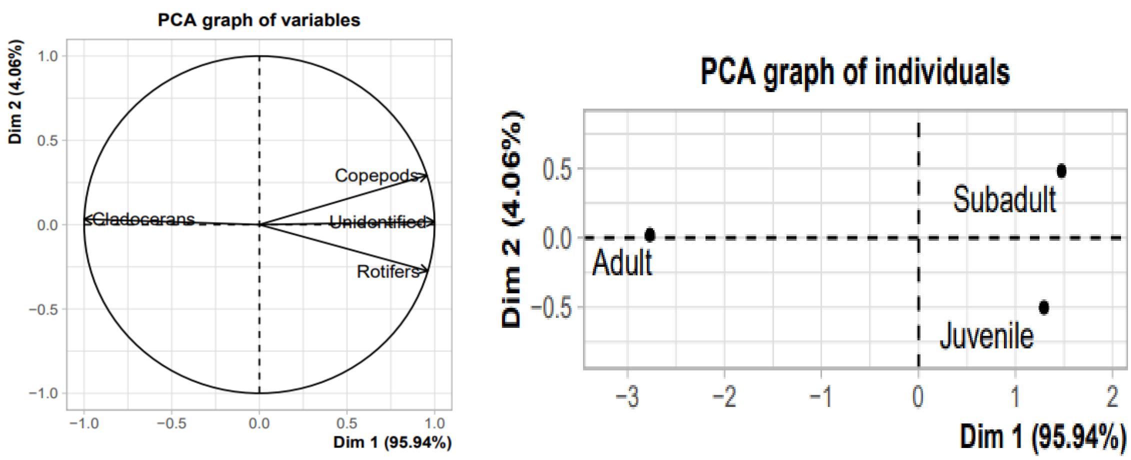

Figure 5: PCA variables graph and PCA individuals graph showing food preference similarities among size classes of *Eucinostomus melanopterus*.

DISCUSSION

Occurrence frequencies enabled to detect that copepod is the most consumed zooplankton by *Ethmalosa fimbriata* with a value of 70.45%, followed by rotifers and cladocerans with respectively 31.81% and 27.84%. It can thus be said that copepod is the preferred prey of this species. The same trend was reported by Gning et al. (2008) in Senegal. Gnohossou et al. (2013) also found that *E. fimbriata* fed on small copepods (Cyclopoids and Harpacticoids) in lake Nokoué. Indeed, the permanent availability of copepods whatever be salinity value in Lake Nokoué (Adandedjan et al., 2017; Chaigneau et al., 2023) could explain the present results.

The peak of zooplankton consumption by *E. fimbriata* is recorded in November with average nutrition index of 5.9. As November is a dry month in Benin, this result is similar to the findings of Faye et al. (2014) who reported that food diet of the species is better during heat periods. Contrarily, the decrease of this index during January with ANI of 1.92, could be explained by the fact that big specimens of *E. fimbriata* were obtained during this month. This hypothesis is granted because the more the species grows, the more its zooplankton consumption decreases. The ANI passes from a value of 6.06 for *E. fimbriata* individuals of 5-7 cm in size to 1.2 for individuals of 13- 15 cm in size. This trend indicates that *E. fimbriata* feeds mainly on zooplankton (micro-organisms) during juvenile stage and consumes large preys when it becomes adult. This could be explained by the fact that big individuals have large choice regarding feed and it confirms the results of Faye et al. (2012) who noticed that in individuals of *E. fimbriata* specimens with fork length around 9.5 cm, copepods are scarce in stomach content.

According to Djidohokpin et al. (2017), big fishes could be able to enlarge their food preference related to the availability of habitual preys. Indeed, the PCA realized in order to make correspondence between size classes and consumed zooplankton species shows preys selectivity trend for size superior to 7 cm. Chronology of results enables to report that *E. fimbriata* has diversified zooplankton diet made of rotifers, cladocerans and mainly copepods that represent its preferred prey. Preys are majorly consumed in November by juveniles.

With an occurrence frequency of 50.54%, cladocerans are the most preferred by *Eucinostomus melanopterus* as prey. It is followed by copepods and rotifers whose occurrence frequencies are respectively 39.01% and 23.07%. Based on results of Chaigneau et al. (2023) showing that cladocerans are less available than copepods and rotifers in Lake Nokoué, it could be concluded that the preference of cladocerans is a free choice. This contrasts with the findings of Gning et al. (2008) in Senegal and Baidoo (2018) in Ghana, who observed that zooplankton preys of *E. melanopterus* are mainly constituted by copepods. This discrepancy can be attributed to differences in the taxonomic composition of zooplankton in the different waterbodies. The results of Baidoo (2018) demonstrated that *E. melanopterus* can exhibit trophic diversification among food components across different waterbodies. Additionally, zooplankton populations are highly dependent on variations in environmental conditions (Adandedjan et al., 2017), leading to fluctuations in the availability of zooplankton prey. Notably, neither study of Gning et al. (2008) and Baidoo (2018) reported cladocerans among the zooplankton prey in their sampling, although the present study indicated that cladocerans are the dominant zooplankton in diet of *E. melanopterus*.

However, prey preference analysis in relation to individual size of *E. melanopterus* reveals that regarding its growth, *E. melanopterus* has specific prey preference during advanced life stages. This is similar to many authors results, like Ramos et al. (2014) and Mofu et al. (2019), who affirmed that in many fish, the diet of the same fish can change with growth with difference between juveniles and adults diet. According to Ramos et al. (2014), this is common for gerreids. The present results suggest that juveniles and sub-adults (individuals ranging from 5-13 cm) of *E. melanopterus* essentially feed on rotifers and copepods. This consumption of copepods during juvenile stages was signaled by Gning et al. (2010) in Sine Salum estuary in Senegal, Ramos et al. (2014) in an Brazilian estuary, de Araújo et al. (2016) in a tropical estuary of northeastern Brazil and Mendonca et al. (2019) in tides pools in Brazil. However, our results are in contrast with others researches about young *E. melanopterus* diet, whose found that young and sub-adults individuals fed mainly on small mollusks larvae) (Gning et al., 2010). According to Qwabe and Cyrus (2021), the diets of the same fish can vary from one estuary to another.

Gning et al. (2010) reported that the diet of *E. melanopterus* individuals depends on prey availability, which is driven by habitat and season variations (Ramos et al., 2014). Adults individuals with size comprised between 13 to 15 cm prefer cladocerans. It is supposed that during its growth especially in immature stages, *E. melanopterus* is not too selective toward zooplankton. Its selectivity toward zooplankton is noticed during advanced growing stages. Indeed, according to Gning et al. (2008), *E. melanopterus* immature individuals' dwell in lagoon before joining sea environment. The study focused essentially on immature individuals of this species. The size and structure of *E. melanopterus* stomach make it challenging to determine the stomach content weight, which is crucial for ANI calculation.

The study takes into consideration a part of the year from September 2021 to January 2022. These months in southern Benin correspond to the short rainy season (September – October) and the long dry season (November – March). It would be advisable to conduct the study across all seasons in southern Benin to gather a more comprehensive dataset. Research works can also be deepened by identifying preys consumed by *E. fimbriata* and *E. melanopterus* using genetic tools. For more comprehension on food diet of these species, it would be important to deepen research through study of reproduction parameters, physiology and ecology in lagoon and marine environment simultaneously for rational stock management.

Conclusion

The current study provided insights into the zooplankton prey of two zooplanktivorous fish species, *Ethmalosa fimbriata* (Clupeidae) and *Eucinostomus melanopterus* (Gerreidae), in Lake Nokoué. The analysis of stomach content revealed that in Lake Nokoué, *Ethmalosa fimbriata* and *Eucinostomus melanopterus* feed on three main zooplankton groups that are copepods, rotifers and cladocerans. *Ethmalosa fimbriata* prefer copepods though *Eucinostomus melanopterus* consumes cladocerans. As soon as size increases, individuals of *Ethmalosa fimbriata* consume low zooplankton and there is no particular difference regarding its zooplankton preference in relation to size. Mature individuals *Eucinostomus melanopterus* prefer cladocerans but there is no strict preference in relation to size gradient.

COMPETING INTERESTS

The authors declare that they have no competing interests.

AUTHORS' CONTRIBUTIONS

HHA, DSDB, GD and ZS contributed to conception of the research project. GD, DSDB, GH, LZ contributed to fish sampling, length measurements and dissection. DSDB and FTO realized zooplankton prey from stomach contents identification. GD and FUD-S conducted statistical analysis. DSDB wrote the original draft. HHA, GD, FUDS, FTO, ZS, SHTZ contributed to the critical review of the article.

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