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RESEARCH ARTICLE ARAŞTIRMA MAKALESİ

Biology, digestive enzymes and organosomatic indices of *Chrysichthys nigrodigitatus* (Lacépède, 1803) from Oyan Dam, Southwestern Nigeria

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Abstract: Some aspects of the biology, digestive enzymes and organosomatic indices of *Chrysichthys nigrodigitatus* purchased from fishermen in Oyan dam was investigated. This study was aimed at providing information on the composition of food materials found in the gut and specific activities of selected enzymes as it affects the domestication of the species. 100 specimens of the species were examined for stomach contents, length-weight relationship, digestive enzyme assay and organosomatic indices, using standard methods. Food items observed were detritus (4%), fish part (12%), Insecta (13%). Sand was observed to be 11% of total stomach volume. The logarithmic equation for length-weight relationship $lnW = 2.68lnl - 3.79$ indicated that an increase in length led to a corresponding increase in weight with 'R' = 0.611, calculated 'r'=0.78, 'a'=2.68 and 'b'=3.79 indicating positive allometric growth pattern. Amylase exhibited high activity in the stomach, while lipase and proteinase in the stomach and posterior intestine. Specific activities of digestive enzymes showed significant differences (p<0.05). Viscerosomatic (2.92± 0.25), hepatosomatic (2.27± 0.22) indices and Fulton condition factor (1.93± 0.06) were recorded. Feed items present in the species confirm its overlapping feeding habit, indicating that the species is an omnivorous detritivore. This was also depicted in the activities of the different digestive enzymes.

Keywords: *Chrysichthys nigrodigitatus*, digestive enzymes, Oyan Dam, length- weight relationship, food item, produce

INTRODUCTION

Fish species of the Claroteidae family has wide commercial value in the Nigeria context (Akinsanya et al., 2007). Of this family, the genus *Chrysichthys* is well commercialized and known as "Inanga" by the people of Ibibio, "Warushe" to the Hausa's, Igangan to the Yoruba's Nigeria. The common silver catfish - *Chrysichthys nigrodigitatus* Lacépède, 1803 is native to Africa and widely distributed in Nigeria's fresh water.

Growth as a function of animal feeding portrays the different metabolic interactions and adjustments that occur within the animal. These are sustained by the nutrient availability and nutritional status of the animal. Studies have shown that the potential of fish to digest its food is highly variable and it's influenced by species, food preference, style of feeding, size, age and temperature (Garcia-Carreno et al., 2002; Gioda et al., 2017). Many problems associated with fish feeding is due to the lack of knowledge of fish species digestive processes (Rønnestad et al., 2013). Investigating the digestive enzymes of a species relate the feeding habits of that fish species to the enzymes that are present in the fish gut. This provides a clue to explaining the digestive processes that take place once food is consumed by the species (Fagbenro et al., 2001).

Several studies have been conducted on the food and feeding habits (Atobatele and Ugwumba, 2011; Kuton and Akinsanya, 2016), length–weight relationship (Kareem et al., 2015; Uneke, 2015) of the species. For successful domestication and culture of any fish species, there is need to investigate the biology of the species. However, there is a paucity of data on the activity of the digestive enzymes of this fish species. This study was aimed at providing information on the composition of food materials found in the gut and specific activities of selected enzymes that break down nutrients in the diets of the species obtained from catches of fishermen in the study area.

MATERIALS AND METHODS

Study Area

The government of Nigeria under the management of the Ogun-Osun River Basin Development Authority (Figure 1) owns Oyan Dam. Its longitudinal and latitudinal location is 3° 16° East and 7° 15° North with an elevation of 43.3 m above sea level on the confluence of Oyan and Ofiki rivers (Ofoezie et al., 1991; O-ORBDA, 1998). The catchment area of the dam is about 9,000 km² within the southern climatic belt of Nigeria. The reservoir is a home for booming fishing activities and fishermen.

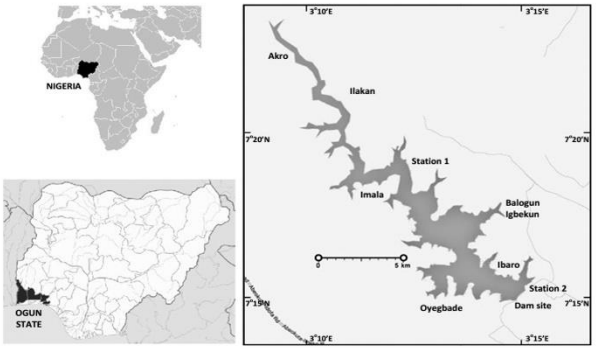

Figure 1. Map showing Oyan Dam (area of study), Ogun State, Nigeria

Collection of Samples

The sampling stations used for the study were the Imala and Ibaro, which are known for their thriving fishing activities (Ikenweiwe et al., 2007). Sampling was carried out every month for five months. On a monthly basis, ten fish samples were purchased from fishermen using gill nets from each location. Conveyance of samples was by the use of an ice chest to the biology laboratory, Department of Zoology, Federal University of Agriculture, Abeokuta for fresh examination.

Samples identification and laboratory Procedure

Species identification was by the guides, as described by Olaosebikan and Raji (2004). The morphometric measure of length was to the nearest 0.1 cm while weight was to the nearest 0.1 g. Standard measuring and digital balance (Camry electronic weighing scale, Model EK3250. Zhongshan Camry Electronic Co, Ltd., Zhongshan, Guangdong, China) were used for length and weight. A pair of stainless–steel scissors and forceps were used to make a longitudinal incision from the anus to the mouth thereby exposing the internal organs of the samples. This were carefully removed using a pair of throngs.

Stomach Fullness Classification

Classification of stomach contents into different categories was as described by Ugwumba and Ugwumba (2007). Categories includes:

0/4 = empty stomach

1/4 = one quarter full stomach

2/4 = half full stomach

3/4 = three quarter full stomach

4/4 = full stomach

Identification of food items in stomach

The stomachs of the experimental fish were cut open and the contents emptied into different petri dishes. Ten percent of normal saline was poured into the petri dish to dislodge clogged particles. Food materials were then graded into various groups and observed under Biobase Optical microscope (BMP-107B). Identification of food items to the species level was done using the key guides described by Mellanby (1965) and Frid (2002). Frequency of occurrence and numerical methods of stomach contents analysis was then used to analyze stomach contents, as described by Adeosun et al. (2017).

For frequency of occurrence, the formula used was:

$$\%\ \text{Occurrence of particular food item} = \frac{\text{No. of point of the particular food item}}{\text{Total number of points of all food}} \times 100$$

Also, according to the description in the Bowen guide for stomach contents identification, the number of stomachs in which the'particular' food particle is found is expressed as a percentage of the total stomachs with food examined.

$$\%\ \text{Occurrence of the i food item in the sample} = \frac{\text{No. of stomachs in which the i item is found}}{\text{No. of stomachs with food in the sample}} \times 100$$

For numerical analysis of stomach content, the expression below was used.

$$\%\ \text{Number of a food item} = \frac{\text{Total number of the particular food item}}{\text{Total number of all food items}} \times 100$$

Length-Weight Relationship

Length-Weight Relationship **(**LWR) was determined as described by Bagenal (1978), using the equation:

Weight of fish = constant 'a' x Length of fish x constant 'b'

$$(W = aL^b) \quad [1]$$

Where: *b* is the slope and *a* is the intercept on the length axis.

Logarithmic transformation of equation 1 gives the straight-line relationship depicted by the equation 2 below:

$$\text{LogW} = \text{Log}a + b\text{LogL} \quad [2]$$

Condition factor (K)

The state of wellness of the samples was calculated using the formula of Froese (2006) as described by Jin et al. (2015).

$$K = 100\ X \frac{W\ (g)}{L^3\ (cm)}$$

W = Weight of fish

L^3= Cube root of standard length of fish

After determining the Fulton's condition factor of the samples, the liver and visceral were removed carefully and weighed using Camry electronic weighing scale (Model EK3250. 5kg). Viscerosomatic index was used as a method of determining the growth performance in fish feeding. The

mathematical equation below was used to calculate organosomatic indices:

$$\text{OSI} = \frac{\text{Weight of organ (g)}}{\text{weight of fish (g)}} \text{ x } 100$$

Gut Enzymes Analysis

Total amylase (α and b) activity

One milliliter of 1% soluble starch in 1/10M sodium acetate buffer pH 5.0 was added to 1 ml of supernatant and the mixture was incubated for 1 hour at 27°C. Two milliliters of DNSA reagent were added to the resultant mixture to terminate enzyme action and the resulting coloured solution allowed to boil for 5 minutes. The solution was then made to 10 ml mark with distilled water and cooled under running tap water. The volume of reducing sugar formed was determined by reading the optical density of the solution at 540 nm against the blank and calculated from a standard curve of Maltose (Swain and Dekker, 1966).

α-Amylase activity

From the supernatant obtained from the total amylase test, 5 ml was heated for 15 minutes at 70°C. After heating, 1 ml of the extract was then incubated in 1 ml of 1% soluble starch in 1/10M sodium acetate buffer solution (pH 5.0) for 1 h at 27°C. At the end, incubation of specific enzyme activity was stopped by the addition of 2 ml DNSA reagent to the reaction mixture. The quantity of reducing sugar formed was then determined, using the same procedure for total amylase activity.

Proteinase activity

The enzyme extract preparation method used for proteinase activity was as for total amylase activity, except that 20 ml of 0.05 M sodium phosphate pH 6.0 was the extracting buffer. The Lowry protein assay method for determining proteinase activity in the enzyme extract was adopted (Lowry et al., 1951). The optical density of the solution was measured at 700 nm against a blank similarly treated (1 ml boiled enzyme extract). Proteinase activity was then calculated using a standard curve of various concentrations of tyrosine (Somkuti and Babel, 1967).

Lipase activity

The method described by Yong and Wood (1977) was adopted for this analysis. Three milliliters of the enzyme extract supernatant was mixed with an emulsion solution that comprised of 100 ml glycerol tributyrate, 4 mg sodium taurocholate, 20 ml 0.1M sodium acetate buffer (pH 5.0) and 10 ml 2.2% $CaCl_2$. The resulting mixture was then incubated for one hour at 40°C. After incubation, 20 ml of absolute ethanol was added and the ensuing solution was titrated against 0.02 M NaOH using a phenolphthalein indicator (0.1 g in 50 ml absolute ethanol and 5 ml water). A blank that contained 3 ml of boiled enzyme extract was similarly treated as the enzyme extract.

Statistical Analysis

All data were subjected to statistical analysis using the Statistical Package for Social Science (SPSS 2007) software. The coefficient of regression was used to assess food preference and style of feeding of the species.

RESULTS

Stomach content

Stomach fullness values for *C. nigrodigitatus* sampled revealed that of the 100 fish specimen sampled, 21% of the stomach were observed to be full while 15% of the stomachs were empty (Figure 2). Insects and scales were found in the majority of the stomach of *C. nigrodigitatus* with food materials. Fish crumbs and nematode were also found in the majority of the guts. Other food items observed were plants, Crustaceans, Rotifers, Arachnida, Mollusca, and detritus (Figure 3)

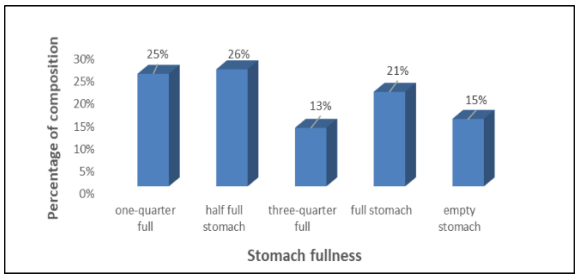

Figure 2. Graphical illustration of stomach fullness

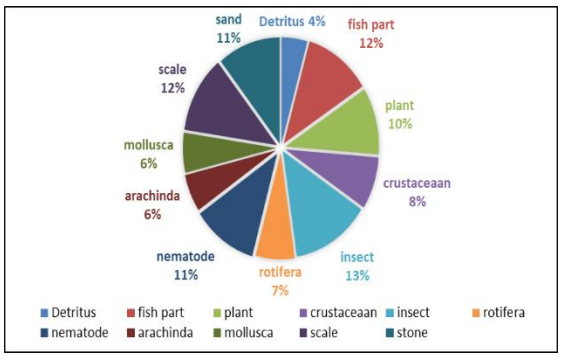

Figure 3. Food items in the stomachs of *Chrysichthys nigrodigitatus*

Length-weight relationship (LWR)

LWR of the species is depicted (Figure 4). The linear equation i.e. $W = 2.68lnl - 3.79$ and the regression parameters are significant F 1.98=153.96 (p<0.05) and correlation (r=0.78). Regression co-efficient R^2 = 0.611.

Organosomatic indices and condition factor

The organosomatic indices and condition factor of *C. nigrodigitatus* are as presented in Table 1. The confidence interval estimation at 99% confidence level using appropriate statistical tool showed that the hepatosomatic (liver somatic) index (HSI) of the sampled specimen was between 2.05 and 2.49 and viscerosomatic index (VSI) between 2.67 and 3.17 with 'K' between 1.87 and 1.99.

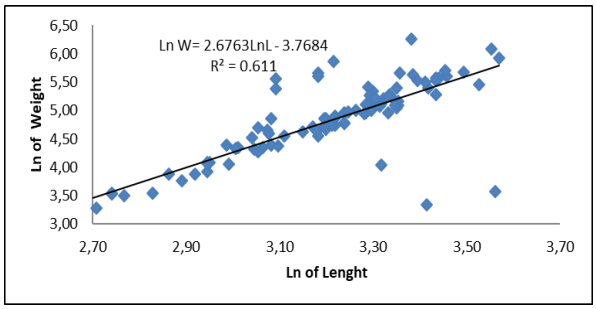

Figure 4. Length-weight relationship of *C. nigrodigitatus*

Table 1. Organosomatic indices and condition factor of *Chrysichthys nigrodigitatus*

Parameters (%)	Mean ± SE	P-value
Liver somatic index	2.27± 0.22	<0.01
Viscerosomatic index	2.92± 0.25	<0.01
Condition Factor	1.93± 0.06	<0.01

Activity of digestive enzymes in the gut of *Chrysichthys nigrodigitatus*

Enzymes activities in the guts are presented in Table 2. Various segments of the guts, showed varying enzymes activity strength. The highest amylase activity was observed in the stomach and was significantly different (p<0.05). Lipase and proteinase activity was lowest in the anterior intestine and increased through to the posterior intestine.

Table 2. Specific activity of digestive enzymes (U/g) in the gut

Digestive enzyme	Anterior intestine	Stomach	Small intestine	Posterior intestine
Amylase	2.29±1.28^{b}	15.69±0.97^{a}	10.54±1.06^{c}	.07±0.99^{d}
Lipase	5.84±1.31^{c}	20.29±1.33	22.62±1.39^{ab}	3.49±3.23^{a}
Proteinase	.57±0.56^{b}	4.15±0.57^{b}	7.01±1.04^{a}	.62±0.96^{a}

Rows with Means ± S.D with superscripts at variance implies statistical difference at 95% probability

DISCUSSION

The composition of the stomach content of the experimental fish is in agreement with the findings of previous studies on this species (Yem et al., 2009; Lawal et al., 2010; Thomas and Ogomade 2019). The study however did not agree with the result presented by Dada and Araoye (2008) who documented higher dominance of plant materials in the stomach of the species from Asa Dam. This could be a result of the low and high abundance of the various food items in the different areas. The prevalence of insect parts observed in this study agreed with the finding of Atobatele and Ugwumba (2011) who posited that *C. nigrodigitatus* consumed more insects as it increased in size with a decrease in preference for the crustacean. However, the study of Udosen and Rufus (2018) reported a higher percentage of phytoplankton in the stomach of the species from Uta-Ewa Creek in the South-South, Nigeria confirming the opportunistic feeding behaviour of the species. The study also confirms the overlapping and omnivorous feeding nature of the species and corroborates the findings of previous researchers (Atobatele and Ugwumba, 2011; Uneke, 2014, Udosen and Rufus, 2018) and could be due to the trophic flexibility and opportunistic feeding behaviour of the species.

The viscerosomatic index, which indicates the growth performance of fish during fish feeding was higher than one. This indicated that the species feeding in the environment during the study was optimal. The high value obtained for the hepatosomatic index also revealed that the species in the study location were in good health condition. This was seen in the value obtained for the Fulton condition factor indicating that the environment is conducive for the survival of the species. The present finding is similar to that documented by Kumar et al. (2010); Lawal et al. (2010). Thomas and Ogomade (2019) also documented similar condition factors for the species. The findings however differ from that of Amoah et al. (2008). The species exhibited a positive allometric growth pattern and corroborated the earlier study by Yem et al. (2009) but disagreed with that of Offem et al. (2008) and Lawal et al. (2010). The correlation value indicated that standard length increased or decreased with a corresponding increase or decrease in weight of *C. nigrodigitatus* in the course of the study which could be linked to food availability and good environmental condition for the survival of the fish species.

The strong specific activity of the amylase enzyme observed in the stomach starting from the anterior intestine is an indication that the breakdown of carbohydrate foods in the fish commenced from the anterior segment (oesophagus) and got stronger in the stomach. This could indicate that the digestion of carbohydrate foods was concluded in these parts of the alimentary canal (AC). A similar result was presented in the study of Odedeyi and Fagbenro, (2010).

The high activity of the lipase enzyme in the AC, especially in the stomach to the posterior intestine could also indicate that lipid metabolism was stronger in the stomach region down to the posterior end of the intestine. A similar observation has been reported by Fagbenro et al., (2000) in *Heterotis niloticus* Cuvier 1829. The omnivorous feeding habit of the species affects the activities of the different enzymes. In another study by Fagbenro et al. (2001) they needed high activity level of proteinase and trypsin in the posterior end of the intestine of *Malapterurus electricus* (Gmelin 1789) which was supported by the findings of this study. The occurrence of proteinase and lipase in the gut of *C. nigrodigitatus* could be associated with the omnivore detritivorous food habits of the species as posited by Tramati et al. (2005).

Chrysichthys nigrodigitatus is an omnivorous detritivore as it exhibited an overlapping feeding habit. The omnivorous feeding habit of the species also affected the activities of the different digestive enzymes. The study provided useful information on the use of the viscerosomatic index as a key for determining the growth performance of fish to fish feeding. The study also concludes that the study area was conducive for the growth and survival of the species.

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AUTHORSHIP CONTRIBUTION

Oghenebrorhie Mavis Oghenochuko, Festus Idowu Adeosun: Conceptualization, idea, design. Fiyinfoluwa Georgina Leramo, Olamide Modinat Adeosun, Paul Bangura: Data curation. Fiyinfoluwa Georgina Leramo: Writing- Original draft preparation. Olamide Modinat Adeosun, Paul Bangura: Revised first draft. Oghenebrorhie Mavis Oghenochuko, Festus Idowu Adeosun: Revised final draft. Oghenebrorhie Mavis Oghenochuko, Festus Idowu Adeosun: Supervision.

CONFLICT OF INTEREST STATEMENT

The authors declare that there are no conflicts of interest

ETHICS APPROVAL

No specific ethical approval was necessary for this study as freshly moribund fish were used for the study.

DATA AVAILABILITY

All relevant data is in the article.

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