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DIGESTIVE ENZYMES IN THE GUT OF THE ELEPHANT SNOUT FISH, *Mormyrus rume* (Valenciennes, 1846) (OSTEICHTHYS: MORMYRIDAE)

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Abstract

Qualitative and quantitative assays were conducted to determine the occurrence and distribution of digestive enzymes in different gut regions of juvenile and adult elephant snout fish, *Mormyrus rume*. Amylase, maltase, lactase, sucrase, chitinase, pepsin, trypsin and lipase were present in the oesophagus, stomach, pyloric caeca, duodenum, ileum and rectum at varying quantities and activities. The wide distribution of enzymes in both juvenile and adult *M. rume* confirms its ability to digest carbohydrate, protein and lipid portions of its diet.

Key words: Digestive enzymes, gut, *Mormyrus rume*.

Introduction

Qualitative and quantitative assays of the enzymes in the gut of a fish contribute toward understanding the nutritional physiology of the fish. The quality of a given diet is directly proportional to its ability to support growth and its nutritional value is determined by the ability of the animal to digest and absorb it (Akintunde, 1985). Tengjaroenkul *et al*. (2000) reported that the distribution and activity of digestive enzymes along the gut change with feeding habits. Tramati *et al*. (2005) noted that the age and/or stage of development influence the anatomical and physiological development of the digestive organs; and the digestive processes correlate with the size and type of food items in fish, thus justifying different dietary habits at various stages of the life cycle (Kuz'mina, *et al.,* 2002). Under natural conditions, adults tend to capture larger prey, which demands a greater digestive effort due to the smaller surface area exposed to enzymatic action. Uys and Hecht (1987) reported that knowledge of the digestive enzymes enhances the development of more efficient diets and rearing techniques.

Major enzymes detected in the gut of tropical fin and shellfishes are proteases, glycosidases and lipases which are responsible for the digestion of proteins, carbohydrates and lipid content of their diets, respectively (Hsu and Wu, 1979; Johnston and Yellowless, 1998; Johnston and Freeman, 2005). Dabrowski and Glogoskwi (1977), Chow and Halver (1980) and Bairagi *et al*. (2002) reported that detritus-inhabiting microflora which produce microbial cellulase imparts the ability to digest cellulose to their host animal. Several studies have been made on the digestive enzymes of different fish species (Olatunde

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and Ogunbiyi, 1977; Uys and Hecht, 1987; Danulat, 1986; Fagbenro *et al.,* 1993, 2000*,* 2001; Ugwumba, 1993; Tramati *et al.,* 2005; Masahiro *et al.,* 2006) but no available information on that of *M. rume*. The objectives of this study are to determine the occurrence and distribution of digestive enzymes in different regions of the gut of juveniles and adults of the elephant snout fish, *M. rume*.

Materials and methods

Forty (40) live *M. rume* were obtained from fishermen in River Ose (Ondo State). Total length, standard length (cm) and weight (g) of each fish sample were taken using a fish measuring board and Ohaus Tripple Balance, respectively. The samples were kept for 48 hours in glass tanks without feeding to bring them to the same physiological conditions as well as ensure the emptiness of their guts. The entire gut (oesophagus to anus) was removed and measured on ice. The gut of each fish was separated into oesophagus, stomach, pyloric caeca, duodenum, ileum and rectum. Tissues from ten specimens each of juveniles and adults fish in wet and dry seasons respectively were pooled and preserved separately in a chest freezer. Each of the regions was weighed after thawing, homogenized in an all-glass homogenizer with ten times its weight of ice-cold neutralized 1% potassium hydroxide to give 1:10 homogenate. The homogenate were centrifuged at 2500 rpm for 20 minutes at 4°C in a refrigerated centrifuge (Eppendorf model 5702R). The precipitant was discarded and the clear supernatant was used as crude enzymes solution without further purification.

Assay of proteases activities

Digestive proteases in the alimentary tract of *M. rume* were determined qualitatively according to the method described by Balogun and Fisher (1970). The reaction mixture for trypsin consisted of 0.5ml of 1% alkaline casein (pH 7.60) and 0.5ml of the enzyme solution. The enzyme solution in the control was heat inactivated in boiling water bath for up to 20minutes. Both the test samples and control was incubated simultaneously for one hour at a constant temperature of 37°C in the water bath (TEMP STAR model). After the incubation, 1% acetic acid solution was added drop by drop to both the control and the test solutions. A change in the test sample compared with the control indicates the presence of enzymes in the test sample. For qualitative determination of pepsin, the reaction mixture consists of 0.5ml of 1% acid casein (pH 2.0) and 0.5ml of enzyme solution. The enzyme extract in the control was denatured by boiling it in a water bath for 20 minutes at 37°C. Both the test samples and the control was incubated simultaneously in an incubator (TEMP STAR model) for two hours. At the end of the incubation period, 1% sodium acetate solution was added drop by drop to each of the sample and control. A change in the test sample compared with the control was taken as an index for the presence of pepsin in the test sample. Usually the presence of trypsin/pepsin was noted by an increase in the turbidity of the test solution.

Quantitative determination of proteolytic activity in the digestive tract of *M. rume* followed the method of Rinderknecht *et al.* (1968). Trypsin was determined quantitatively in a reaction mixture consisting 1ml 10mg egg-albumin solution, 2ml phosphate buffer (pH 8.0) and 0.5ml enzyme solution. The enzyme in the control was boiled at 37°C for 20 minutes in a water bath. The reaction

mixture for the determination of pepsin was as that of trypsin but the phosphate buffer for the pepsin was pH 2.0. Both the test sample and the control were incubated for an hour at 100°C. During incubation period, there was regular shaking of the tubes to ensure uniform reaction. At the end of the incubation period 3ml of ice-cold phosphate buffer was added to the test tubes. The mixture was filtered immediately through filter paper Whatman No. 1 and the absorbance of the filtrate was determined using digital colorimeter (Lab-Tech Model). The blank was used to adjust the colorimeter to zero.

Preparation of Calibration Curve for protein

The stock used consists of 100µg/ml of egg-albumin that is, 10mg of egg-albumin dissolved in 100ml of water. Preparation of serial dilutions was made for stock solution ranging from 20 -100µg/ml of egg-albumin. Each concentration was made in duplicate tubes; with 1ml of each dilution in different test tubes and 3ml of biuret was added to all the test tubes. The egg-albumin in the blank tube was replaced with distilled water. Both the test samples and control was incubated for ten minutes at 37°C in a water bath for colour development. The optimal density of the solution was determined colorimetrically at 540nm. The result obtained from these was used in the preparation of a calibration curve for determination of protein using egg-albumin. From the curve absorbance was converted to enzyme activity using the formula:

$$\text{Unit of enzyme activity} = \frac{\text{amount of amino acid released in µg/ml}}{\text{Time}}$$

Assay of glycosidases activities

Glycosidases activities in the digestive tract of *M. rume* were determined qualitatively using the substrates such as starch, maltose, lactose, sucrose, chitin and cellulose. The enzymes were detected according to the methods described by Olatunde *et al.* (1988). Each assay mixture consisted of 0.4ml of 1% substrate, 0.2ml phosphate buffer (pH 8.0) and 0.1ml of enzyme solution. All the experiments were in triplicates. The enzyme extract for the control was heat-inactivated for 20 minutes in boiling water bath prior its addition to the reaction mixture. The tubes containing the test and the control mixture were incubated for 3minutes at room temperature. The hydrolysis of polysaccharides and non-reducing disaccharide were determined qualitatively in terms of the appearance of reducing properties using Benedicts test. 5.0ml of Benedict's qualitative reagent was added to both the test sample and control then heated in a water bath at 100^{o}C for one hour. The appearance of a brick red to cream yellow precipitate was taken as an indication of positive reaction.

Glycosidases were determined quantitatively by the dinitrosalicylate (DNS) methods described by Plummer (1978). The presence of free reducing sugars was initially determined and the amount of DNS reduced in the presence of appropriate substrate and extracts were compared. The substrates used for the assays are starch, maltose, sucrose, lactose, chitin and cellulose. The reaction mixture consisted of 0.4ml of 1% substrates, 0.2ml phosphate buffer pH 8.0 and 1.6ml DNS and 0.1ml of enzyme extract. The enzyme extract in the control was replaced with distilled water. The control and test samples were allowed to stand at room temperature for three minutes, then 1.0ml

(DNS) was added to each and placed in boiling water for five minutes. They were removed and left to cool for 30 minutes, after which 1.8ml distilled water was added to make up to 4.0ml (for dilution) and mixed properly. The amount of reducing sugar produced on enzymatic reaction was estimated colorimetrically using digital colorimeter, (Lab-Tec Model) at 540mm. The blank mixture was used to adjust the colorimeter to zero**.**

Preparation of calibration curve for glucose

The stock solution used for the preparation of calibration curve of glucose consisted of 0.1g D-glucose per 100ml (i.e. 1mg/ml glucose). Serial dilutions ranging from 0 to 0.8mg/ml were prepared from the stock. This was made up to 1ml by addition of distilled water. A blank tube that contains 1ml of distilled water was used to adjust the colorimeter to zero. Each concentration was prepared in duplicate. To all the test tubes 1.6ml of alkaline 3.5-DNS reagent was added and mixed properly. They were placed in a boiling water bath for 10mins. 1ml of each of the diluted stock was taken into another test tube and further diluted with addition of 1ml distilled water. The absorbance of the reaction mixture was read at 540nm against a blank containing buffer without enzyme on the digital colorimeter.

$$\text{Unit of enzyme activity} = \frac{\text{amount of glucose released in mg/ml}}{\text{Time}}$$

Assay of Lipases activities

Lipolytic activity was determined at 37°C as described by Ogunbiyi and Okon (1976). The reaction mixture was made of 1.0ml of 25% olive oil emulsion pH 7.0 and 0.2ml of enzyme extract. Sample test and the control were incubated for one hour at 37°C in a water bath, after which 3.0ml of 95% ethanol and two drops of phenolphthalein were added to each test tube including the control. The reaction mixture was titrated against 0.05M NaOH to a similar pink colour. An increase in the titre value of test sample was compared with the control to confirm the presence of lipase in the test sample. Quantitative assay of lipase followed the same method for qualitative analysis and the titre value was read as lipase activity for each of the gut regions.

Statistical analysis

All data obtained from the qualitative and quantitative analyses were analyzed using means and standard deviation for qualitative analysis while regression analysis and correlation coefficient were determined in the quantitative estimation. Data were also subjected to analysis of variance (ANOVA) with season (wet and dry), size (juveniles and adults) and gut region (oesophagus, stomach, pyloric caeca, duodenum, ileum and rectum) as sources of variation.

Results

Lipases

The qualitative assay of digestive enzymes in the gut of *M. rume* juveniles and adults are presented in Table 1. The results showed that lipase activity was present with different strengths along the entire gut regions of both the juvenile and adult *M. rume.* Lipase activity was very strong in the duodenum, strong in the ileum and pyloric caeca but weak in the oesophagus, stomach and rectum of both the juveniles and adults *M. rume* in both seasons. The quantitative analyses of lipase in the gut of *M. rume* are presented in Tables 2 and 3 for juveniles and adults in wet and dry seasons, respectively. The highest lipolytic activity was recorded

in the duodenum and was significantly different ($p<0.05$) from the other regions while the least activity was recorded in the oesophagus. Lipase activity was higher in the adults than the juveniles while there was no significant difference ($p>0.05$) between seasons.

Proteases

Pepsin was present in the stomach, pyloric caeca and duodenum and was strong in the stomach of both the juveniles and adults but weak in pyloric caeca and duodenum and absent in oesophagus, ileum and rectum (Table 1). Trypsin was present in the duodenum and ileum and strong in the ileum. The presence of proteases was not detected at all in the oesophagus and rectum of *M. rume* juveniles and adults in both seasons. It was observed that high activity of proteases occurred in the stomach and ileum than other gut regions. Pepsin activity was very high in the stomach and low in the pyloric caeca and duodenum, there was significant difference ($p<0.05$) in activity between juveniles and adults. Trypsin activity was high in the ileum and low in the duodenum and were significantly different ($p<0.05$) between juveniles and adults. Proteolytic activity was not recorded in the oesophagus and rectum of both juveniles and adults *M. rume* in both seasons.

Glycosidases

All the six glycosidases assayed (α-amylase, maltase, lactase, sucrase, chitinase, cellulase) were present in all the gut sections except the rectum of *M. rume* (Table 1). Also, α-amylase and maltase were present in all the gut regions except the rectum. α-amylase was very strong in both pyloric caeca and duodenum, strong in the stomach while the other glycosidases detected were weak in the various gut regions. Chitinase and cellulase were detected in the stomach, pyloric caeca and the duodenum of juveniles and adults. There were variability of glycosidases activities in different gut regions of juveniles and adults in wet and dry seasons. The highest activity was in the pyloric caeca and duodenum and the least activity occurred in the oesophagus (Tables 2 and 3). There were significant differences ($p<0.05$) between activity of enzymes in both stages but no significant difference ($p>0.05$) between seasons.

Table 1. Digestive enzymes activities in the gut of *M. rume*

	Oesophagus	Stomach	Pyloric caeca	Duodenum	Ileum	Rectum
Lipases	+	+	++	+++	++	+
Pepsin	–	++	+	+	–	–
Trypsin	–	–	–	+	++	–
α-Amylase	+	+	++	++	+	–
Maltase	+	+	+	+	+	–
Lactase	–	+	+	+	+	–
Sucrase	–	+	+	+	+	–
Chitinase	–	++	+	+	–	–
Cellulase	–	+	+	+	–	–

+++ very strong enzyme activity, ++ strong enzyme activity, + weak enzyme activity, - no enzyme activity

Table 2. Enzyme activities in the gut of juveniles and adults of *M. rume* from River Ose (Wet season)

	Juveniles								
	Proteases[2]			Glycosidases[3]					
Gut regions	Lipases[1]	Pepsin	Trypsin	α-amylase	Maltase	Lactase	Sucrase	Cellulase	Chitinase
Oesophagus	$0.830\pm0.15^{a**}$	0	0	$0.07\pm0.006^{a*}$	$0.017\pm0.006^{a*}$	0	0	0	0
Stomach	$2.310\pm0150^{b*}$	$0.153\pm0.020^{a*}$	0	$0.140\pm0.010^{b*}$	$0.090\pm0^{b*}$	$0.073\pm0.006^{a*}$	$0.067\pm0.006^{a*}$	$0.020\pm0.010^{a*}$	$0.033\pm0.006^{a*}$
Pyloric caeca	$4.207\pm0.600^{c*}$	$0.100\pm0^{b*}$	0	$0.503\pm0.02^{c*}$	$0.093\pm0.006^{c*}$	$0.080\pm0.017^{b*}$	$0.047\pm0.006^{b*}$	$0.030\pm0.0^{b*}$	$0.033\pm0.006^{a*}$
Duodenum	$6.580\pm0.53^{d*}$	$0.103\pm0.012^{b*}$	$0.070\pm0.53^{a*}$	$0.347\pm0.032^{d*}$	$0.063\pm0.006^{d*}$	$0.053\pm0.021^{c*}$	$0.033\pm0.012^{c*}$	$0.017\pm0.006^{c*}$	$0.017\pm0.006^{b*}$
Ileum	$4.847\pm0.064^{e*}$	0	$0.170\pm0.010^{b*}$	$0.033\pm0.006^{e*}$	$0.013\pm0.006^{e*}$	$0.023\pm0.006^{d*}$	$0.017\pm0.006^{d*}$	0	0
Rectum	$1.207\pm0.012^{f*}$	0	0	0	0	0	0	0	0
Adults									
Oesophagus	$0.900\pm0.10^{a**}$	0	0	$0.107\pm0.006^{a**}$	$0.107\pm0.006^{a**}$	0	0	0	0
Stomach	$3.540\pm0.399^{b**}$	$0.183\pm0.015^{a**}$	$0.340\pm0.182^{b**}$	$0.090\pm0^{b**}$	$0.083\pm0.006^{a**}$	$0.083\pm0.015^{a**}$	$0.020\pm0^{a**}$	$0.050\pm0^{a**}$	$0.340\pm0.182^{b**}$
Pyloriccaeca	$4.833\pm0.533^{c**}$	$0.103\pm0.006^{b**}$	0	$0.547\pm0.021^{c**}$	$0.130\pm0.035^{c**}$	$0.060\pm0.010^{b**}$	$0.073\pm0.006^{b**}$	$0.037\pm0.015^{b**}$	$0.050\pm0.017^{a**}$
Duodenum	$7.200\pm0.100^{d**}$	$0.107\pm0.006^{c**}$	$0.103\pm0.012^{a**}$	$0.393\pm0.023^{d**}$	$0.070\pm0.020^{d**}$	$0.047\pm0.015^{c**}$	$0.047\pm0.012^{c**}$	$0.040\pm0.010^{c**}$	$0.037\pm0.006^{b**}$
Ileum	$5.257\pm0.137^{e**}$	0	$0.187\pm0.021^{b**}$	$0.037\pm0.006^{e**}$	$0.033\pm0.006^{e**}$	$0.033\pm0.006^{d**}$	$0.023\pm0.006^{d**}$	0	0
Rectum	0	$2.050\pm0.218^{f**}$	0	0	0	0	0	0	0

a, b, c, d, e, f = for each enzyme, data along the same column with different superscript alphabets are significantly different ($p<0.05$)

*,** = for each enzyme, data along the same row with different asteriks are significantly different ($p<0.05$)

[1] milliequivalents of fatty acids/mg protein at 37°C, [2] change in optical density at 595 nm/hr/mg of L-tyrosine/hr at 37°C, [3] mg glucose/min/mg protein at 37°C

Table 3. Enzyme activities in the gut of juveniles and adults of *M. rume* from River Ose (dry season)

	Juveniles								
	Proteases			Glycosidases					
Gut regions	Lipase	Pepsin	Trypsin	α-amylase	Maltase	Lactase	Sucrase	Cellulase	Chitinase
Oesophagus	$0.843\pm0.002^{a*}$	0	0	$0.013\pm0.006^{a*}$	$0.010\pm0^{a*}$	0	0	0	0
Stomach	$2.350\pm0.150^{b*}$	$0.150\pm0.017^{a*}$	0	$0.140\pm0.026^{b*}$	$0.93\pm0.006^{b*}$	$0.070\pm0.010^{a*}$	$0.070\pm0.010^{a*}$	$0.017\pm0.006^{a*}$	$0.033\pm0.006^{a*}$
Pyloric caeca	$5.03\pm0.170^{c*}$	$0.100\pm0^{b*}$	0	$0.490\pm0.026^{c*}$	$0.083\pm0.015^{C*}$	$0.073\pm0.006^{a*}$	$0.057\pm0.006^{b*}$	$0.037\pm0.006^{b*}$	$0.023\pm0.006^{b*}$
Duodenum	$6.700\pm0.265^{d*}$	$0.100\pm0^{b*}$	$0.083\pm0.006^{a*}$	$0.370\pm0.026^{d*}$	$0.063\pm0.006^{d*}$	$0.040\pm0.010^{b*}$	$0.037\pm0.006^{c*}$	$0.020\pm0^{c*}$	$0.020\pm0.0^{c*}$
Ileum	$5.23\pm0.076^{e*}$	0	$0.150\pm0.020^{b*}$	$0.033\pm0.015^{e*}$	$0.027\pm0.015^{e*}$	$0.023\pm0.006^{c*}$	$0.013\pm0.006^{d*}$	0	0
Rectum	$1.383\pm0.189^{f*}$	0	0	0	0	0	0	0	0
	Adults								
Oesophagus	$0.897\pm0.09^{a**}$	0	0	$0.103\pm0.006^{a**}$	$0.013\pm0.006^{a**}$	0	0	0	0
Stomach	$3.47\pm0.329^{b**}$	$0.187\pm0.012^{a**}$	0	$0.333\pm0.211^{b**}$	$0.103\pm0.012^{b**}$	$0.080\pm0.010^{a**}$	$0.080\pm0.010^{a**}$	$0.017\pm0.006^{a**}$	$0.050\pm0^{a**}$
Pyloric caeca	$5.37\pm0.38^{c**}$	$0.113\pm0.006^{b**}$	0	$0.510\pm0.030^{c**}$	$0.133\pm0.012^{c**}$	$0.067\pm0.006^{b*}$	$0.070\pm0.010^{b**}$	$0.052\pm0.017^{b**}$	$0.060\pm0^{b**}$
Duodenum	$7.257\pm0.129^{d*}$	$0.110\pm0.010^{b**}$	$0.100\pm0.010^{a**}$	$0.380\pm0.020^{d**}$	$0.070\pm0.017^{d**}$	$0.043\pm0.015^{c**}$	$0.050\pm0.010^{c**}$	$0.033\pm0.012^{c**}$	$0.050\pm0.010^{a*}$
Ileum	$5.287\pm0.141^{e**}$	0	$0.180\pm0^{b**}$	$0.037\pm0.012^{e**}$	$0.027\pm.0.012^{e*}$	$0.030\pm0.010^{d*}$	$0.013\pm0.006^{d**}$	0	0
Rectum	$3.767\pm0.195^{f**}$	0	0	0	0	0	0	0	0

a, b, c, d, e, f = for each enzyme, data along the same column with different superscript alphabets are significantly different ($p<0.05$)

*,** = for each enzyme, data along the same row with different asteriks are significantly different ($p<0.05$)

[1] milliequivalents of fatty acids/mg protein at 37°C, [2] change in optical density at 595 nm/hr/mg of L-tyrosine/hr at 37°C, [3] mg glucose/min/mg protein at 37°C

Discussion

Qualitative and quantitative analyses of glycosidases showed that *M. rume* is capable of digesting carbohydrates in its diet. The presence of high glycosidase activity is an indication that the fish is well equipped for digestion the carbohydrates consumed. Such high level of glycosidase activity implies the ability of *M. rume* to utilize carbohydrate food component as an energy source. The presence of cellulase in the gut of *M. rume* in this study agrees with Fagbenro *et al.* (1993) who noted that occurrence of cellulase in the gut of *H. bidorsalis* as a

rather unusual development because it occurs in few vertebrates, and that where it occurs, the source is usually traced to the micro flora inhabiting their guts. Tramati *et al.* (2005) also reported cellulase activities in the stomach, pyloric caeca, foregut, midgut and hindgut of *Diplodus puntazzo* juveniles and adults. The digestion of starch appears to start from the oesophagus and continues up to the ileum. The high glycosidase activity in the pyloric caeca and duodenum ensure complete digestion of carbohydrates in these regions. The presence of lactase was also reported by Fagbenro *et al.* (1993) in *Heterobranchus bidorsalis* and Lagler *et al*. (1977) in the pyloric caeca of the trout, though lactase is known to be associated with milk digestion in mammals.

The strong activities of chitinase in the stomach of the adult and average activities in the pyloric caeca and duodenum could be due to dietary habits of the adult fish which has insects as the prominent food item (Ugwumba *et al*., 1990; Ipinjolu *et al*., 2005; Odedeyi *et al*., 2006). This agrees with Fagbenro *et al*. (2000) who recorded high chitinase activity in both stomach and duodenum of the electric catfish (*Malapterurus electricus*) and associated this to its chitin-eating habit (i.e. feeding on crustaceans and insects). Similarly high chitinase activities have been detected in the stomachs and intestines of Atlantic cod, fed on whole crustaceans (Danulat, 1986). Lindsay (1984) reported that the primary function of gastric chitinase in fish is likely to disrupt chemically the chitin envelope of the prey. However, chitin (a major structural component of the cuticle of insects and the exoskeleton of crustaceans) like cellulose, has no utilizable energy value to fish (Fagbenro *et al*., 2001).

Digestion of protein in *M. rume* starts in the stomach. The absence of proteases in the oesophagus showed that this region only serve as a place of passage of food materials to the stomach while the rectum serve as a temporary storage organ for undigested food materials or waste before they are voided from the body (Olatunde and Ogunbiyi, 1977; Olatunde *et al*., 1988). The high peptic activities recorded in the stomach of *M. rume* suggest the consumption of protein-rich diet by this species. High peptic activity, possibly pepsinogen (an inactive pepsin precursor which eliminates the risk of self digestion of the stomach) that was recorded in *M. rume* agrees with Fagbenro *et al*. (2001) who reported high peptic in the stomach of the electric catfish. Smith (1980) reported that peptic activity in fish occurs in acid conditions (pH 2.0-4.0) as in higher vertebrates, and is about 150 times greater than that of mammalian pepsin in its affinity for its substrate (Ananichev, 1959).

The occurrence of high trypsin activity, an alkaline protease, in the ileum of *M. rume* agrees with Fagbenro *et al*. (2001) who also recorded high trypsin activity in the ileum of the electric catfish. Olatunde and Ogunbiyi (1977) attributed high trypsin activities in *Schilbe mystus* to the large amount of fish and insect materials in the diet of the species. Pepsin activity was high in both juveniles and adults of *M. rume.* This does not agree with Qian and Qian (1998) who observed that trypsin activity was higher than that of pepsin in mandarin fish, *Siniperca chuatsi.* Wang *et al*. (2002) reported that protease activities became higher with growth of the fish, *Paralichthys olivaceus*, which was similarly observed in this study. High pepsin and trypsin activities

recorded in this study can be attributed to *M. rume* consuming protein-rich diets, similarly with *H. bidorsalis* (Fagbenro *et al.*, 1993, 2001) and tropical catfishes, *Physailia pellucida, Eutropius niloticus* and *S. mystus* (Olatunde and Ogunbiyi, 1977). Protein digestion was initiated by pepsin and completed by trypsin; Uys and Hecht (1987) and Fagbenro *et al*. (1993) reported that partial hydrolysis of protein is subsequently completed by the combined action of trypsin and chymotrypsin when the food reaches the intestine. Proteolytic activity was not influenced by season as the diet of the fish remained the same from season to season (Odedeyi *et al*., 2006).

Lipase was detected in all the regions of the gut in *M. rume;* indicating a uniform distribution in the entire gut system. This observation is similar to that reported for *Heterotis niloticus* by Fagbenro *et al*. (2000). Swarup and Goel (1975) also observed lipases along the entire gut of some teleosts. The least lipolytic activity observed in the oesophagus and was more abundant in the pyloric caeca, duodenum and ileum which are the neutral and alkaline regions of the tract. This agrees with Tramati *et al.* (2005) who reported that lipase was more abundant in the neutral-alkaline gut regions of juvenile and adult *D. puntazzo.* Fagbenro *et al*. (1993) reported that lipase activity was average and restricted to the posterior regions of the gut of *H. bidorsalis* while Olatunde and Ogunbiyi (1977) reported that lipase activities were not detected in the gut of *P. pellucida, E. niloticus* and *S. mystus* and remarked that this observation was a surprise in view of the fact that the diets of *E. niloticus* and *S. mystus* include clupeid, a fish which contains plenty of fats.

The presence or absence of certain digestive enzyme in individual animal as reported by Buddington (1985) depends on its diet and functional morphology of the various part of the gut. Hence, glycosidases, proteases and lipases in the gut of *M. rume,* a detrivore is in correlation with its diet. This shows the ability of *M. rume* to utilize a wide range of nutrients in its environment effectively, which is one of the qualities of a fish with aquaculture potential.

In this study, levels of digestive enzymes activities in the adults were higher than those measured in the juveniles, which agrees with Tramati *et al.* (2005) that the digestive processes of *D. puntazzo* correlates with the size and composition of food. Lee *et al*. (1984) and Francois *et al.* (2000) also recorded that the activities of glycolytic enzymes increased with fish mass.

Results from this study suggest that the gut of both juveniles and adults of *M. rume* are fortified with the major digestive enzymes (glycosidases, proteases, lipases) capable of digesting carbohydrate, protein and lipid contents of its diet. Amylase, maltase, lactase, sucrase, chitinase, pepsin, trypsin and lipase were present in the oesophagus, stomach, pyloric caeca, duodenum, ileum and rectum at varying activities.

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