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Original Research Article

A Comparative Histology and Histochemistry of Stomach of Three Teleostean Fishes

B. Laxma Reddy* and G. Benarjee

Fisheries Research Lab, Department of Zoology, Kakatiya University, Warangal-506 009, India

**Corresponding author.*

Abstract

In fresh water teleosts structure of stomach is quite similar to that of oesophagus except the modifications of the lining of the epithelium into characteristic gastric epithelium and gastric glands. Stomach is formed by four layers that is mucosa with superficial epithelium and glandular epithelium, submucosa, muscularis and serosa. In the present study histomorphological and histochemical changes have been observed in the stomach of three fresh water teleost fish species, *Labeo rohita*, *Wallago attu* and *Clarias batrachus*.

Keywords

Clarias batrachus

Histochemistry

Histomorphology

Labeo rohita

Stomach

Wallago attu

Introduction

Fish exhibits diverse nature of dietary habits the histomorphological adaptations of the alimentary canal to the specific food habit of the species concerned become inevitable. In fact, each fish species has its own structural adaptations of the alimentary canal towards its specific food habit, which vary greatly in regard to the ratio of animal and plant materials ingested. Fishes are generally classified as herbivores, carnivores and omnivores. Some fishes are grouped as specialists like planktivores, insectivores, sponge feeders, crustacean feeders, mollusk feeders, algal fish feeders and piscivores eating flesh form prey, scales and fins. In the present study an attempt has been made to characterize comparatively, the histomorphology and histochemistry of the stomach in three different fishes- herbivorous *Labeo rohita*, carnivorous *Wallago attu* and omnivorous *Clarias batrachus*.

Materials and methods

For the present investigation fresh water teleosts, *Labeo rohita*, *Wallago attu*, *Clarias batrachus* were collected from fishermen at nearby from water tanks and were sacrificed by cervical dislocation. The stomach was removed and cut into small pieces then they preserved in various fixatives i.e., Bouin's, Susa, Carnoy and Zenker's fluid. After the routine histological preparation of the tissue, the sections of 5micron thickness were cut on rotatory microtome. To study the normal histology of the stomach Heidenhains Azan (Gurr, 1962) stain was used. Different kinds of histochemical procedures were also followed to elucidate the chemical nature of stomach. The presence of carbohydrates, proteins, and lipids and nucleic acids was determined by using the technique mentioned by Pearse (1968), Gomorie (1952), McManus and Mowry (1960), Lillie (1965), Humason (1967) and Bancroft (1975).

Results and discussion

In the herbivorous *Labeo rohita* the stomach is not present. The anterior part of the intestine is known as intestinal bulb or intestinal swelling. This is a special feature in these fishes. The intestinal bulb of *Labeo rohita* is a thick walled wide structure extending below the air bladder. It is anterior broader cardiac parts into which opens dorsally by the pancreatic and bile ducts, and a posterior narrower pyloric part without pyloric caeca, which are common in other teleost fishes. In the carnivorous fish *Wallago attu*, the oesophagus dilates into true stomach. It is short pear shaped, thick walled and secular lies on the ventral side of the airbladder slightly towards the left side of the visceral cavity. It consists of three main region, the proximal cardiac, middle pyloric and the distal blind fundus region. The structure of the stomach is similar to that of oesophagus, but the modification of the lining of the stomach into characteristic gastric epithelium and gastric glands distinguishes it from the oesophagus, in which the mucous folds are thin.

The stomach in the omnivorous *Clarias batrachus* is demarcated externally from the oesophagus by its large rounded structure. Stomach is also distinguished from the oesophagus internally by the differences in the mucosal folds which are thin in oesophagus and become thicker and wavy in out line in the stomach. The stomach in this fish is wider in the middle than at the two ends and looks like a sac. Its narrow distal end continues into the intestine and the junction is marked by a distinct variation. In the present study indicates that stomach is quite similar to that of oesophagus except the modifications of the epithelium into characteristic gastric epithelium and gastric glands. The transition from the cardiac stomach to the pyloric stomach is gradual. The mucosal lining of the cardiac stomach is raised into straight short folds. These folds become thick and conspicuous in the pyloric region. The stomach of this fish is also formed by the same layers as found in other two fishes in the present study. The stomach in fresh water teleosts exhibits species variation (Sarkar, 1959; Khanna, 1961; Saxena and Bakshi, 1964; Agarwal and Sharma, 1966; Dalela, 1969; Moitra and Ray, 1977, 1979; Ray and Moitra, 1982 and Ghosh and Das, 1987).

In the teleostean fishes, a well developed stomach has formed to receive and store the injested food material. The structure of the stomach is quite similar to that of oesophagus except that the epithelial lining of the stomach has formed into the characteristic gastric epithelium and gastric glands. The passing of the oesophagus into the stomach is recognized by distinct histological variations. The deep longitudinal folds of the oesophagus become reduced and gastric glands begin to appear. This function of oesophagus and stomach is referred as “Oestogaster” by Kamal Pasha (1963). It is formed by four, layers that is mucosa with superficial epithelium and glandular epithelium, submucosa, musularis and serosa. The columnar cells of the gastric epithelium absorptive in function (Greene, 1912; Dawes, 1929; Blake, 1936). The mucous secreting protects the surface of the stomach from the mechanical injury. The cells of cardiac region were believed to help in the digestion of food and the mucous cells of the pyloric region in the neutralization of the acidity of gastric juice before it enters in to the intestine.

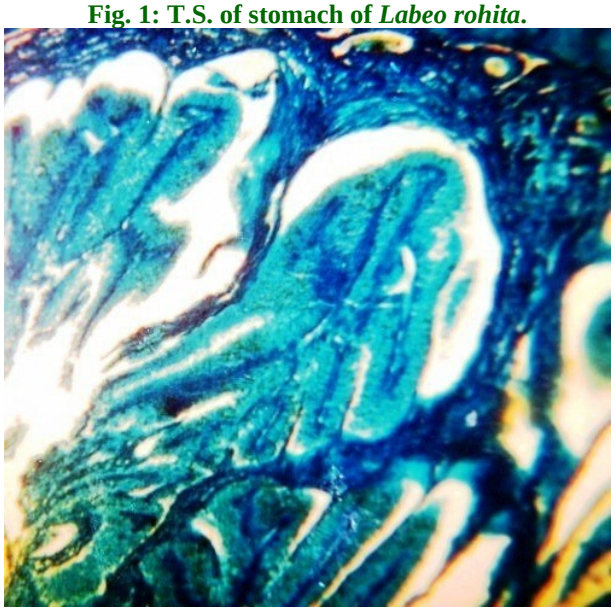
The histochemical tests conducted on the stomach of three fishes *Labeo rohita*, *Wallago attu* and *Clarias batrachus* demonstrated the presence of varied amounts of carbohydrates, proteins and lipids. The transverse sections of the stomach of all the three fish species are depicted in Figs. 1, 2, and 3. The identification of carbohydrates and carbohydrate containing groups has made by using the standard technique Periodic acid/ Schiff reaction. The response to PAS reaction in *Labeo rohita* was moderate, strong in *Wallago attu* and in the case of *Clarias batrachus*. The striated border of the stomach was intensively positive while the other layers are moderately stained. These variations in the intensity of staining indicated variable concentrations of carbohydrates in different fishes. The variations in the amount of carbohydrates are due to the types of the food fish consumed. In all the three above fishes, other tests have also been conducted to determine the specificity of carbohydrate nature, because PAS alone does not visualize all the carbohydrate groups. The positivity with PAS may be due to glycogen, free aldehydes and mucopolysaccharides. Therefore, for the presence of glycogen in the tissues, PAS/Saliva was used in the technique. The PAS reaction was found to be resistant to saliva digestion suggesting the absence of glycogen. The PAS reaction was blocked in the acetylation solution and was restored after deacetylation showing the presence of 1 : 2 glycol groups. Similarly PAS alone was employed, without oxidizing the sections in periodic acid to show the presence of free aldehydes.

The sections of stomach when subjected to alcian blue at 1.0 PH, the intensity of reaction in *Labeo rohita* is mild

one, because of the ill developed mucosal folds containing mucous secreting cells. This further indicates less quantity of sulfated mucosubstances. But in the case of *Wallago attu*, alcian blue at both pH levels responded suggesting the presence of sulphated mucosubstances, hyaluronic acid, sialomucins and weak mucosubstances. The same technique when applied on *Clarias batrachus* stomach the intensity was much more, suggesting large amounts of mucosubstances in the walls of the stomach. Alcian blue at 2.5pH followed by PAS revealed that the mucous cells of the stomach in these fishes stained bluish purple colour indicating the presence of a mixture of both acid and neutral mucins. The gastric cells were positive to PAS indicating the occurrence of neutral mucins.

The PAS positive reaction seen on the surface of the gastric epithelial cells indicated an absorptive function in stomach. The reports of other authors (Greene, 1912; Blake, 1910) indicated that some absorption particularly of fats occurs in teleostean stomach. The positivity of PAS also suggests that the occurrence of glycol groups and aldehydes. The mucoid nature of gastric columnar cells has been reported for different fish species by employing histological and histochemicl methods (Kapoor, 1953; Kamal pasha, 1963; Agarwal and Sharma, 1966; Tandon and Sharma, 1968; Khanna and Mehrotra, 1969; Moitra and Ray, 1977 and 1979).

Fig. 1: T.S. of stomach of *Labeo rohita*.



All the layers of the stomach have responded moderate to intense with Bromophenol blue suggesting that the occurrence of variable quantities of basic proteins. In *Labeo rohita* the distribution of protein content was observed uniformly in all the layers of stomach. While in *Wallago attu* the muscular and submucosa were more positive, the gastric glands exhibited little intensity. On the other hand *Clarias batrachus* showed that striated border of the stomach was intensely positive.

However the intensity of reaction varied. The stomach layers where also positive to Millons reaction and ferric ferry cyanide suggesting the occurrence of tyrosine containing proteins and sulfhydryl groups of proteins. When KMno4/AB was employed the presence of disulphides was indicated. The occurrence of protein bound amino groups was found with Ninhydrin/Schiff. Similarly congored stain demonstrated the presence of glycoproteins in the stomach of all three species of fishes.

Fig. 2: T.S. of stomach of *Wallago attu*.

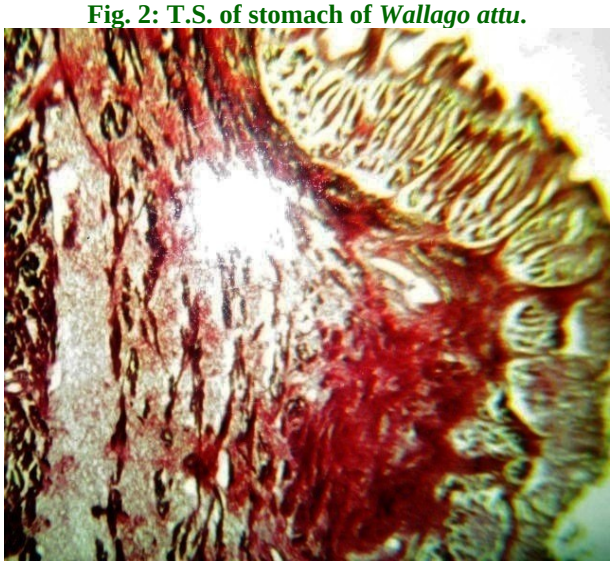


Fig. 3: T.S. of stomach of *Clarius batrachus*.

Besides the occurrence of various carbohydrates and different proteins, the stomach of the above three fishes elucidated the lipid nature of the stomach walls. The presence of lipids was shown by Sudan Black B, while copperpthalocyanin demonstrated the occurrence of phospholipids.

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