

CHAPTER - FOUR

HISTOLOGICAL AND HISTOCHEMICAL OBSERVATIONS ON MUCOSUBSTANCES IN THE STOMACH (CARDIAC AND PYLORIC) OF THE CHAMAELEON ZEYLANICUS LAURETI AND DISCUSSION

STOMACH

The stomach was a thick walled, swollen, tubular part of alimentary canal. Broadly it was divisible into three regions : an anterior cardiac, middle fundic and posterior pyloric region. The oesophagus opened in the cardiac region while the pyloric stomach continued to the duodenum. The histological and histochemical observations on proximal and distal regions of the stomach are described hereafter.

OBSERVATIONS

I) CARDIAC STOMACH :

A) HISTOLOGICAL OBSERVATIONS :

The H-E stained preparations (Fig.10) revealed the usual four coats : mucosa, submucosa, muscularis and serosa in the wall of cardiac stomach. The muscularis was thick walled than the oesophagus (Fig. 14).

I) Mucosa :

It was folded. Folds were numerous and broad. The folds were projected into the lumen. The folds were broad and sometimes they called gastric rugae. Single layered epithelium lined the mucosa and the epithelial cells were tall columnar type. Goblet cells were not observed. The well defined cardiac glands were observed below the epithelial lining which were formed by invagination of epithelial cells. The gland cells were of two types, the mucous neck cells

Table 3 : Histochemical reactivities of mucosubstances in the cardiac stomach of C. zeylanicus laureti.

Sr. No.	Histochemical reactions	Epithelial cells	Mucosa		Submucosa	Muscularis	Serosa
			Mucous neck cells	Cardiac glands			
				Oxyntic cells			
1	PAS	++++P	+++±P	-	+P	+±P	+±P
2	Ph - PAS	+P	-	-	-	-	-
3	D - PAS	++++P	+++±P	-	+P	±P	+±P
4	AB pH 1.0	+B	-	-	-	-	-
5	AB pH 1.0 - PAS	++++P	+++±P	-	+P	+±P	+±P
6	AB pH 2.5	+B	-	-	-	-	-
7	AB pH 2.5-PAS	++++P	+++±P	-	+P	+±P	+±P
8	C. I.	+B	-	-	-	-	-
9	C.I. - PAS	++++P	+++±P	-	+P	+±P	+±P
10	AF	+P	-	-	-	-	-
11	AF - AB pH 2.5	+P	-	-	-	-	-
12	Azure A pH 1.5	+0	+0	-	-	-	-
13	Azure A pH 3.0	+0	+0	-	±0	+0	+0
14	Azure A pH 4.5	++M	+++±0	-	+0	+±0	+±0
15	Sulfation Azure A pH 1.5	++M	+++±M	-	+M	+±M	+±M
16	CEC + 0.1 M Mg ⁺⁺	-	-	-	-	-	-
17	CEC + 0.2 M Mg ⁺⁺	+B	-	-	-	-	-
18	CEC + 0.4 M Mg ⁺⁺	±B	-	-	-	-	-
19	CEC + 0.6 M Mg ⁺⁺	-	-	-	-	-	-
20	M - 37°C	+B	-	-	-	-	-
21	DM - 37°C	+B	-	-	-	-	-
22	M - 60°C	-	-	-	-	-	-
23	DM - 60°C	-	-	-	-	-	-
24	Acid hydrolysis-AB pH 2.5	+B	-	-	-	-	-
25	Sialidase-AB pH 2.5	+B	-	-	-	-	-
26	Hyaluronidase-AB pH 2.5	+B	-	-	-	-	-
27	Pepsin - AB pH 2.5	+B	-	-	-	-	-

(Figs. 12, 14) and the oxyntic cells. These glands observed opened into the gastric pits.

ii) Submucosa :

As in oesophagus the submucosa consisted of connective tissue. It was extended into the gastric folds and was provided with blood vessels and nerve fibres.

iii) Muscularis :

Submucosa was followed by thick layered muscularis. The muscle layer distinguished into an inner circular and an outer longitudinal muscles. The inner circular muscle layer was thicker than the outer longitudinal muscle layer.

iv) Serosa :

It was the outer coat formed of thin cellular membrane contributed by the visceral peritoneum.

In both the sexes of chameleons the same histological structure was observed.

B) HISTOCHEMICAL OBSERVATIONS

The histochemical reactivities of mucosubstances in various histological sites of the cardiac stomach are recorded in Table No.3 and the distribution of mucosubstances is shown in photomicrographs (Figs. 10 to 18).

I) Mucosa :

i) Epithelial Cells :

The epithelial cells in the cardiac stomach exhibited intense PAS reactivity (Figs. 11, 12) which could be blocked by phenylhydrazine pretreatment (Fig. 13) (it was retained but slightly) indicated the presence of neutral mucosubstances and acidic mucosubstances. However, the diastase or α -amylase digestion (Fig.14) had no effect on the staining intensity of PAS which indicated the absence of glycogen in these cells.

The presence of neutral mucosubstances in these cells was also inferred from the blue-purple staining with AB pH 1.0 - PAS (Fig.17) AB pH 2.5 - PAS (Fig.18) and C.I.-PAS sequential staining as well as metachromatic pink staining with Azure A at pH 4.5 and Azure A at pH 1.5 following sulfation.

Poor alcianophilia with AB pH 1.0 (Fig.15) which remained unaltered at pH 2.5 (Fig.16) indicated the presence of acidic mucosubstances which were sulfomucins but absence of carboxymucins in these cells. The presence of sulfomucins in these cells was also characterized by blue-purple staining with AB pH 1.0 - PAS (Fig.17), poor purple staining with AF alone and with AF-AB pH 2.5 combined histochemical procedures. These sulfomucins were resistant to mild methylation but active methylation abolished the alcianophilia which failed to restore even after saponification procedure. These sulfomucins were hyaluronidase resistant and there was no enhancement in the alcianophilia by prior pepsin digestion.

Thus, the above mentioned histochemical reactivities lead to the conclusion that the epithelial cells of cardiac stomach contained neutral mucins (predominant) and the poor quantities of sulfomucins.

ii) Cardiac Glands :

Two types of cells have been identified in the cardiac glands—mucous neck cells and oxyntic cells.

a) Mucous Neck Cells :

The cells in the glands, showed moderate to intense PAS reaction (Figs. 11,12) which was resistant to diastase and α -amylase digestion (Fig. 14) but could completely be blocked by prior phenylhydrazine treatment indicating the absence of glycogen but presence of neutral mucosubstances in these cells.

The negative staining of these gland cells with AB pH 1.0 (Fig. 15), AB pH 2.5 and C.I. (Fig. 16) and AF revealed absence of acidic mucosubstances. This was also confirmed by pepsin digestion which did not exhibit any alcianophilia. The presence of only neutral mucosubstances in these gland cells was also substantiated by their only PAS reactivity (moderate to intense, magenta colouration) in sequential staining procedures such as AB pH 1.0 – PAS (Fig.17), AB pH 2.5 – PAS (Fig. 18) AND C.I. – PAS and metachromatic staining with azure only after sulfation of the sections.

The aforementioned histochemical observations revealed the presence of only neutral mucins in the gland cells of cardiac stomach.

b) Oxyntic Cells :

These cells in the cardiac glands remained unstained with PAS (Figs. 10,11), AB pH 1.0 (Fig.15), AB pH 2.5, C.I. (Fig. 16), AF and Azure A. Pepsin digestion did not modify these results. Therefore it was concluded that these cells did not contain neutral or acidic mucosubstances.

II) Submucosa :

The connective tissue in this layer of cardiac stomach exhibited poor PAS staining (Figs. 11,12). This PAS reactivity was remained unchanged with diastase or α -amylase digestion indicated the absence of glycogen. However, the PAS reactivity could completely be blocked by phenylhydrazine pretreatment (Fig.13). This result indicated the presence of only neutral mucosubstances in the connective tissue of this layer. Moreover, the connective tissue remained unstained with AB pH 1.0 (Fig. 15), AB pH 2.5, C.I. (Fig.16) and AF and exhibited only orthochromatic blue staining with Azure A at all pH levels and metachromatic pink staining with Azure A at pH 1.5 only after sulfation. These results indicated the presence of neutral mucosubstances in the connective tissue of the submucosa of the cardiac stomach.

III) Muscularis :

The muscularis of the cardiac stomach exhibited weak PAS staining reactivity (Figs. 11,12). This PAS reactivity was partially blocked by diastase or α -amylase digestion (Fig.14) indicated the

presence of glycogen. The PAS reactivity could completely be blocked by phenylhydrazine pretreatment (Fig.13). This result indicated the presence of only neutral mucins but absence of acidic mucosubstances in this layer. This layer exhibited negative staining with AB pH 1.0 (Fig.15), AB pH 2.5, C.I. (Fig.16) and AF or AF-AB pH 2.5 only purple staining with AB pH 1.0-PAS (Fig.17), AB pH 2.5 (Fig.18) and C.I.-PAS, only orthochromatic blue staining with azure A at and above pH 1.5 and metachromatic pink staining with azure A at pH 1.5 only after sulfation.

Thus, these results indicated the presence of only neutral mucins (weak) and glycogen in the muscularis of cardiac stomach.

IV) Serosa :

The serosa layer of the cardiac stomach showed histochemical results (Figs. 11 to 18) similar to the connective tissue of sub-mucosa of this organ except that the PAS reactivity was weak. Therefore, it was concluded that the serosa of cardiac stomach also contained only neutral mucosubstances (weak).

II PYLORIC STOMACH

It was the distal part of stomach which was narrower and tubular than the cardiac stomach.

A) HISTOLOGICAL OBSERVATIONS :

H-E stained sections of this part (Figs. 19,20) also revealed usual four coats such as mucosa, submucosa, muscularis and serosa.

i) Mucosa :

It was folded but folds were few in numbers. The folds were broad which were provided with numerous secondary folds. The mucosal epithelium was single layered formed mainly of tall columnar cells and few goblet cells. The lamina propria was more prominent in this region than the cardiac stomach. Numerous glands were observed, which were of three types. The differentiation of glands was very clear in histochemical observations particularly in combined histochemical procedures such as AB pH 1.0 - PAS (Fig.24), AB pH 2.5-PAS (Fig. 25) and C.I. - PAS (Fig. 26). The glands showed only one type of cells (Fig.20) referred to them as mucous cells.

ii) Submucosa :

The submucosal connective tissue layer was, thin, nonglandular and was projected into mucosal folds. It was provided with blood vessels and nerve fibres.

iii) Muscularis :

It was thick and distinguished into an inner circular muscle layer and an outer longitudinal muscle layer. This layer was particularly thick towards the distal part of pyloric stomach forming the pyloric sphincter.

iv) Serosa :

It was the thin outermost layer surrounding the muscularis.

B) HISTOCHEMICAL OBSERVATIONS :

The histochemical observations in the different histological sites of the pyloric stomach are recorded in Table No.4 and the distribution of mucosubstances are illustrated in photomicrographs (Figs. 21-27).

I) Mucosa :

i) Epithelial Cells :

Numerically these cells were more than the goblet cells in the pyloric stomach. These cells exhibited intense PAS staining (Fig. 21) which could be blocked with phenylhydrazine pretreatment (it was retained but slightly) indicated the presence of both the neutral mucosubstances and the acidic mucins. The PAS reactivity was not changed with diastase or α -amylase digestion (Fig. 22) indicated the absence of glycogen.

These cells exhibited negative staining with AB pH 1.0 and the alcianophilia appeared at AB pH 2.5 and C.I. (Fig. 23). These results indicated the presence of carboxymucins but absence of sulfomucins. The absence of sulfomucins and the presence of carboxymucins was also inferred from negative staining with AF and only blue staining with Af - AB pH 2.5 sequential staining. These observations were also supported by only purple staining with AB pH 1.0 - PAS (Fig. 25) and complete loss of alcianophilia at mild as well as active methylation and subsequent restoration of alcianophilia with both the methylations followed by saponification, loss of alcianophilic staining after acid hydrolysis and sialidase digestion.

Table 4 : Histochemical reactivities of mucosubstances in the pyloric stomach of C. zeylanicus laureti.

Sr. No.	Histochemical Reactions	Mucosa					Sub-mucosa	Muscularis	Serosa
		Ciliated epithelial cells	Goblet cells	Glands Type-I	Glands Type-II				
					Type-I cells	Type-II cells			
1	PAS	++++P	+++P	++++P	++++P	++++P	+P	+P	+P
2	Ph - PAS	+P	+++P	-	++++P	-	±P	±P	±P
3	D - PAS	++++P	+++P	++++P	++++P	++++P	+P	±P	+P
4	AB pH 1.0	-	+++B	-	++++B	-	±B	±B	±B
5	AB pH 1.0 - PAS	++++P	+++P	++++P	++++B	++++P	+BP	+BP	+BP
6	AB pH 2.5	+B	+++B	-	++++B	-	±B	±B	±B
7	AB pH 2.5-PAS	++++BP	+++B	++++P	++++B	++++P	±BP	±BP	±BP
8	C. I.	+B	+++B	-	++++B	-	±B	±B	±B
9	C.I. - PAS	++++BP	+++B	++++P	++++B	++++P	±BP	±BP	±BP
10	AF	-	+++P	-	++++P	-	±P	±P	±P
11	AF - AB pH 2.5	+B	+++P	-	++++P	-	±P	±P	±P
12	Azure A pH 1.5	-	+++M	+O	++++M	+O	±M	±M	±M
13	Azure A pH 3.0	+M	+++M	+O	++++M	+O	±M	±M	±M
14	Azure A pH 4.5	+M	+++M	++++O	++++M	++++O	±M	±M	±M
15	Sulfation Azure A pH 1.5	+M	+++M	++++M	++++M	++++M	±M	±M	±M
16	CEC + 0.1 M Mg ⁺⁺	+B	+++B	-	++++B	-	±B	±B	±B
17	CEC + 0.2 M Mg ⁺⁺	-	+++B	-	++++B	-	±B	±B	±B
18	CEC + 0.4 M Mg ⁺⁺	-	+++B	-	++++B	-	±B	±B	±B
19	CEC + 0.6 M Mg ⁺⁺	-	+++B	-	++++B	-	±B	±B	±B
20	M - 37°C	-	+++B	-	++++B	-	±B	±B	±B
21	DM - 37°C	+B	+++B	-	++++B	-	±B	±B	±B
22	M - 60°C	-	-	-	-	-	-	-	-
23	DM - 60°C	+B	-	-	-	-	-	-	-
24	Acid hydrolysis-AB pH 2.5	-	+++B	-	++++B	-	±B	±B	±B
25	Stalidase-AB pH 2.5	-	+++B	-	++++B	-	±B	±B	±B
26	Hyaluronidase-AB pH 2.5	+B	+++B	-	++++B	-	±B	±B	±B
27	Pepsin - AB pH 2.5	+B	+++B	-	++++B	-	±B	±B	±B

These carboxymucins were hyaluronidase resistant. These were also resistant to pepsin digestion.

The presence of neutral mucins in these cells was also inferred from pink staining with AB pH 1.0 - PAS (Fig. 24), blue purple staining with AB pH 2.5 - PAS (Fig. 25) and C.I. - PAS (Fig. 26) sequential staining procedures and enhanced metachromasia with azure A at pH 1.5 followed by saponification.

Thus the aforementioned histochemical observations revealed the presence of neutral mucosubstances (moderate) and sialomucins (poor).

ii) Goblet Cells :

These cells were few in number which exhibited moderate PAS reactivity. The PAS reactivity was resistant to phenylhydrazine pretreatment and diastase or α -amylase digestion (Fig. 22) indicating the absence of neutral mucosubstances and glycogen.

These cells reacted moderately towards AB at pH 1.0 and similar moderate blue staining was also seen in this layer with AB pH 2.5 and C.I. (Fig. 23). Moreover, these cells remained only moderately blue in sequential staining procedure such as AB pH 1.0 - PAS, (Fig. 24), AB pH 2.5 - PAS (Fig. 25). Therefore, it was concluded that the goblet cells in these cells contained only sulfomucins.

The above conclusion that the goblet cells contained only sulfomucins was further supported by only moderate purple staining with AF alone or with AB pH 2.5 step afterwards, moderate metachromasia with azure A even at lower pH (pH 1.5) and persistent alciano-

philic staining in CEC technique, in presence of graded concentration of Mg^{++} . Weak alcianophilia persisted even with 0.8 M Mg^{++} concentration. The sulfomucins were resistant to mild methylation. The active methylation abolished the alcianophilia of these cells and the subsequent saponification could not reverse it, which also confirmed the presence of sulfomucins. The sulfomucins were resistant to hyaluronidase digestion.

The aforementioned results therefore, indicated the presence of only sulfomucins in these cells of pyloric stomach.

iii) Glands :

Three types of glands have been identified in pyloric stomach depending upon the type of mucosubstances secreted by the gland cells, which were glands-type I, glands-type II and glands-type III.

a) Glands- Type I :

The cells of this type of glands showed intense PAS reactivity which was completely blocked by phenylhydrazine pretreatment, however, the diastase digestion or α -amylase digestion had no effect on the intensity of PAS in these cells. These initial results indicated the presence of only neutral mucosubstances while absence of glycogen.

The negative reaction of these cells with AB pH 1.0, AB pH 2.5, C.I. (Fig. 23) and AF revealed absence of acidic mucosubstances. This was also confirmed by pepsin digestion which did not exhibit any alcianophilia.

The presence of neutral mucosubstances in these cells was also substantiated by their only PAS reactivity (intense magenta colouration) in sequential staining procedures such as AB pH 1.0 - PAS (Fig. 24), AB pH 2.5 - PAS (Fig. 25) and C.I. - PAS (Fig. 26) and metachromatic staining with Azure A at pH 1.5 only after the sulfation of the sections.

Thus, the aforementioned histochemical observations revealed the presence of only neutral mucosubstances (intense) in these cells of glands-type I.

b) Glands- Type II :

The cells of this type of glands exhibited an intense PAS reactivity. The PAS reactivity was resistant to prior phenylhydrazine treatment and diastase or α -amylase digestions, indicating the absence of neutral mucosubstances and glycogen.

These cells reacted intensely towards AB pH 1.0 and similar intense blue staining was also seen in these cells with AB pH 2.5 and C.I. (Fig. 23). Moreover, these cells remained only intense blue in sequential staining procedures such as AB pH 1.0 - PAS (Fig. 24), AB pH 2.5 - PAS (Fig. 25). Therefore, it was concluded that these cells contained only sulfomucins.

The above conclusion that these cells contained only sulfomucins was further supported by only intense purple staining with AF alone or with AB pH 2.5 step afterwards, intense metachromasia with azure A even at lower pH (pH 1.5) and persistent alcianophilic

staining in CEC technique in presence of graded concentration of Mg^{++} . Weak alcianophilia persisted even with 0.8 M Mg^{++} concentration. The sulfomucins were resistant to mild methylation, however, active methylation abolished alcianophilia of these cells and the subsequent saponification could not restore it, which also confirmed the presence of sulfomucins. The sulfomucins were resistant to hyaluronic acid.

Thus, the aforementioned results indicated the presence of only sulfomucins in these cells of glands-type II.

c) Glands- Type III :

In these glands two types of cells were observed : the type-I and the type-II cells.

i) Type - I Cells :

These cells exhibited the histochemical results which were practically identical to the gland cells observed in glands-type I. Therefore, it was concluded that these type-I cells in these glands contained only neutral mucosubstances.

ii) Type - II Cells :

These type of cells exhibited the results identical to the cells of glands-type II. Therefore, it was concluded that these cells contained only sulfomucins (intense).

II) Submucosa :

The connective tissue of submucosa showed only poor staining with PAS (Fig.21) which could partially be blocked by phenylhydrazine

pretreatment but diastase digestion (Fig. 22) could not alter the PAS staining. These observations indicated that the connective tissue in this layer contained trace amount of both the neutral and acidic mucosubstances but the absence of glycogen.

The connective tissue showed trace alcianophilia at pH 2.5 and C.I. (Fig. 23), indicating the presence of only sulfomucins (trace). The sulfomucins in the connective tissue was also confirmed from only trace purple staining with AF staining alone or with AF - AB pH 2.5 step, metachromasia with azure A at pH 4.5 and after sulfation, loss of alcianophilia by active methylation which could not be restored by subsequent saponification and persistent alcianophilia in the presence of 0.2 M Mg^{++} conc in CEC technique. The sulfomucins were hyaluronidase resistant. Similarly, pepsin digestion had no effect on the intensity of alcianophilia staining with AB pH 2.5. These sulfomucins were resistant to acid hydrolysis and sialidase digestion.

The fact that the connective tissue of submucosa also contained trace amount of neutral mucosubstances were inferred from blue-purple staining with AB pH 1.0-PAS (Fig. 24), AB pH 2.5-PAS (Fig. 25) and C.I.-PAS (Fig. 26).

Therefore, it was concluded that the connective tissue in submucosa of pyloric stomach contained the neutral mucosubstances (trace) and sulfomucins (trace).

III) Muscularis :

The muscularis of pyloric stomach exhibited identical histochemical reactivities (Figs. 21-27) to that described for connective tissue of submucosa. The only difference was that this layer showed the presence of glycogen as the PAS reactivity get diminished with diastase or α -amylase digestion (Fig.22).

Therefore, it was concluded that this layer contained the neutral mucins (trace), sulfomucins (trace) and glycogen.

IV) Serosa :

This layer showed practically identical histochemical results which were observed in the connective tissue of the submucosa of this organ. Therefore, it was concluded that the serosa of pyloric stomach contained the mixture of neutral mucins (trace) and sulfomucins (trace).

DISCUSSION

CARDIAC STOMACH :

The histological observations on the cardiac stomach of the chamaeleon revealed typical four layers- mucosa, submucosa, muscularis and serosa in the wall. The mucosa was lined with single layer of epithelial cells which were tall columnar. The gastric glands containing mucous neck cells and oxyntic cells were observed below the epithelial lining layer. The gastric mucosa of the reptiles has been investigated by Ludicke (1963), Ferri et al. (1974a,b), Ferri et al. (1975), Amer and Ismail (1976), Loo and Swan (1978), Giroud et al. (1979), Suganuma et al. (1981a), Mandlik (1983), Jadhav (1985)

and Patil (1987) which resembles to some extent to the present observations. Ferri et al. (1974b) described some cells clearly in the gastric mucosa of snake, to which Helander (1981) described as mucous neck cells.

The present investigation revealed the gastric glands containing two types of cells mucous neck cells and oxyntic cells and these glands observed opened into gastric pits. Similarly, Patt and Patt (1969) described the presence of mucous neck cells and oxyntic cells in the gastric glands of reptiles. These two types of cells have also been reported by Mandlik (1983) in the gastric glands of the Indian skink; by Jadhav (1985) in turtle and ground lizard and by Patil (1987) in five species of reptiles he has studied.

Thiruvathukal (1965) identified parietal, chief and mucous cells in gastric glands of the turtle, C. picta. Ferri et al. (1975) described the presence of surface epithelial cells and gastric glands with neck cells and body cells in stomach of a snake, X. marremii. Patt and Patt (1969) described that in mammalian stomach HCl is elaborated by parietal cells while pepsin is the secretion of chief cells. However, in amphibians and reptilian stomach the same cells elaborated both the HCl and pepsin, These cells were described as oxyntic cells (Amer and Ismail, 1976) or oxynticopeptic cells (Giroud et al., 1979). The submucosa, mucosa, muscularis and serosa in the chamaeleon resembled to these layers in other reptilian stomach described by Patt and Patt (1969), in Indian skink described by Mandlik (1983), in ground lizard as described by Jadhav (1985) and in five species of reptiles as described by Patil (1987).

The mucosubstances present in the various layers from mucosa to serosa of cardiac stomach resemble to the typical mucins present in other tissue and cells. Atypical mucosubstances are absent in the cardiac stomach of the chamaeleon.

The histological and histochemical results in the present investigation showed the absence of sex dimorphism in the cardiac stomach of the skink.

The histochemical results under present investigation revealed the presence of neutral mucosubstances and sulfomucins in epithelial cells of cardiac stomach of chamaeleon. Such type of epithelial cells of cardiac stomach have variously been described as lining epithelial cells, columnar mucous cells surface mucous cells, pavement epitheliocytes, covering epithelial cells and surface goblet cells. Similar histochemical study by Giroud et al. (1979) in the lizard, I. scincoides and by Helander (1981) in other few reptiles demonstrated the PAS positive granules in the surface mucous cells, the surface mucous cells have been reported to contain only neutral mucosubstances in the stomach of Indian skink (Mandlik, 1983) in turtle and ground lizard (Jadhav, 1985) and in wall lizard, chamaeleon and varanus (Patil, 1987). On the other hand Loo and Swan (1978) reported that the lining epithelial cells contain neutral and acidic mucins in the Australian lizard. Mogil'naya et al. (1978) also found neutral carbohydrates and sialomucins in these cells of some reptiles.

The mucous neck cells contained PAS positive granules as described by Helander (1981). Similar results are reported for these cells in lizards (Loo and Swan, 1978; Mandlik, 1983; Jadhav, 1985 and Patil 1987) and in a snake (Ferri et al., 1975). The results in the present investigation revealed the presence of only neutral mucins in the gland. On the other hand Jadhav (1985) found neutral mucosubstances and AB unreactive but AF reactive mucosubstances in the mucous cells of turtle. In the present investigation, however, atypical sulfomucins were not found.

Oxyntic cells in the present investigation remained unstained with all histochemical techniques even with PAS indicated the absence of any mucosubstances in these cells. However, Helander (1981) described that these cells in reptiles stain lightly with PAS. Similarly Patil (1987) reported the presence of neutral mucosubstances in the oxynticopeptic cells of varanus and snake (poor) and of turtle, wall lizard and chamaeleon (weak) depending on their respective poor and weak only PAS reactivities. But, Mandlik (1983) in his investigation reported that the oxyntic cells present in the cardiac stomach of M. carinata remain unstained by all histochemical techniques indicated the absence of any mucosubstances in these cells. The results in the present investigation, in this way matched with the results obtained by Mandlik.

The connective, muscularis and serosa of cardiac stomach under present investigation contained only neutral mucosubstances, and glycogen was present only in muscularis. Mandlik (1983) also reported that these layers contained only neutral mucins and glycogen in muscularis of Indian skink.

PYLORIC STOMACH :

In general the pyloric stomach consists of mucosa to serosa layers with broad muscular folds lined with single layer of epithelium. The mucosal epithelium contains columnar epithelial cells and goblet cells. The present investigation also reveals three types of glands depending upon the nature of mucosubstances elaborated by them. These glands possess only one type of cells unlike the glands present in cardiac stomach. The epithelial cells are described as pyloric cells in the stomach of a snake, X. merremii (Ferri et al., 1975). Giroud et al. (1979) described that in the lizard I. scincoides the gastric pits lead into glands that are extremely coiled in the proximal stomach but becomes progressively shorter and straight in the distal stomach. Mandlik (1983) stated that the mucosal epithelium contains only goblet cells, and pyloric glands also possess only one type of cells. Jadhav (1985) also stated that the mucosal epithelium consists of goblet cells in ground lizard and columnar cells in turtle. He also added that the short and alveolar glands are present in both these reptiles.

The remaining layers viz. submucosa, muscularis and serosa in chamaeleon resembled to the respective layers in the other reptilian stomach described by Patt and Patt (1969), in the skink described by Mandlik (1983), in the turtle and ground lizards as stated by Jadhav (1985) and as suggested by Patil (1987). Histological observations stated that there is no sexual dimorphism in the chamaeleon.

The mucosubstances characterised in the pyloric stomach of the lizard under present investigation were histochemically identical to similar mucosubstances in the other tissues and cells. Sexual dimorphism is also absent in the pyloric stomach of chamaeleon under present investigation.

Regarding the distribution of mucosubstances in the pyloric stomach of the chamaeleon, the present investigation showed the presence of moderate amount of neutral mucosubstances and poor quantities sialomucins in the epithelial cells. However, among the other reptiles studied, Giroud et al. (1979) reported on only PAS reactive mucous cells in the entire mucosa of stomach in a lizard, T. scincoides, Ferri et al. (1975) also identified neutral mucins in the pyloric cells of stomach of the snake, X. merremii, Mandlik (1983) reported that in the pyloric stomach of skink present only the goblet cells which secrete only the neutral mucosubstances. Similarly, Jadhav (1985) and Patil (1987) also reported that the pyloric stomach contains only mucous cells secreting only neutral mucins in the reptiles they have studied.

In this way the present histochemical studies showed slightly different results for the epithelial cells than that have been reported earlier in other reptiles by previous authors.

In the present investigation three types of glands are observed. Histochemically these glands differ from each other some are with broad lamina and some are with short lamina. El-Taib and Jarrer (1983) reported that the glands are short and alveolar in turtle,

M. caspica. Similarly, Jadhav (1985) described about the short and alveolar glands in the turtle and ground lizard. On the other hand Suganuma et al. (1981a) described that the distal portions of the stomach is devoid of glandular structures in skink, E. latiscutatus, but Mandlik (1983) reported pyloric glands in Indian skink, M. carinata. Patil (1987) reported two types of glands in the pyloric stomach of snake. Pyloric glands in the present investigation show some similarities and variations with the existing literature. In general the cells in the glands - type I under present investigation secrete only neutral mucins. The cells in the pyloric glands contain only neutral mucosubstances in Indian skink (Mandlik, 1983), in ground lizard (Jadhav, 1985); in turtle, chamaeleon, varanus and snake (Patil, 1987).

The cells in the glands- type II in chamaeleon under present investigation secrete only sulfomucins while, the glands-type III contain two types of cells which secrete both neutral mucins and sulfomucins. Similar histochemical results have been stated by number of previous authors. Suganuma et al. (1981a) found some of the glands in the pyloric stomach of turtle containing neutral mucosubstances and sialomucins. Jadhav (1985) identified neutral, sulfo and sialomucins in the pyloric glands of turtle. On the other hand Patil (1987) investigated two types of cells in the pyloric glands of turtle, some of them containing a mixture of neutral mucosubstances and sialomucins. According to him the pyloric glands in snake contain two types of glands, the glands with wider lumina

contain neutral mucosubstances predominant and those with narrow lumina contain only weak quantities of neutral mucosubstances.

The connective tissue in the submucosa, muscularis and serosa of the cardiac stomach in the present investigation contain neutral (trace) sulfomucins (trace) besides the glycogen present only in muscularis. Similar histochemical results have also been obtained by Mandlik (1983) in Indian skink, M. carinata.