DISCUSSIONS

DISCUSSION

Mollusca have been the favorite of collectors and skilled amature naturalists, and today they form the experimental material of physiologists, neurologists, geniticist and and bischemists. They are easily caught, patient of observation and wonderfully diversed in their adaptation. Available literature give us very little knowledge about the physiological significance of mucins in the different organ systems of these animals as compared to the their number distribution and disposition The present inverstigation of mucosubstances in the digestive tract of <u>Cypraea</u> arebica arebica was thought out for the purpose, to get an insight about the probable nature of histology and histochemistry and a possible role in the physiology of the digestion. Ghe following is the discussion regarding the different organs of digestive tract of the imarine snail Cypraea arebica arebica .

BUCCAL MASS

In general the prosobranch mouth leads to buccal cavity lined internally by cuticle and wovered by the inner layer of cilary epithelium which contains mucocytes. Beneath this layer there in circular muscle layer. posteriorly the cavity continues as buccal mass having complex assemblage of muscles supports and operates the redula. (Hyman, 1967). In a histochemical work of mucus secretary cells in the epithellial lining of

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alimentary tract in general in Marisa cornuarietis, shows 6 types of cells. They are distingwished by the morphological observation and are related with 4 different types of mucosubstances. (Demian et al., 1972). Several unicellular glands are seen in buccal wall (Martoja, et al, 1976) Carbohydrate digestion in the snail Tegula funebrails has been studied and amylase, laminarase and alginase are reported. (Berrie et al., 1967). In Cellana rediala redula helps for scapping the rock surface. (Rao 1975). He divides the tract into foregut midgut and hindgut. Amylase was detected in foregut. Peculiar feeding was observed in <u>Maxwellia</u> santorosana the mollus in aquarium drilled a tapered hole into upper valve of pelcypod choma arcana. Drilling lasted for 9 days. (Wicksten, 1980) Contribution for redula, its morphology, mechanism, orientation is comparitively immense in mallusc literature. Bibliographic discription of redula of about 100 publications has listed by Schilder (1971). External anatomy of Cypraea fulfoni and C.capensis with redular morphology has been discussed (Kilburn and Donald, 1973). Notes on redular teeth of 7 species of Indonesia are descibed and discussed painting out the variations as regards to structure and form. (Somedihardjo, 1973). Possible functional evolutionary trend is traced by studing the fundamental aduptive features of rhiphidiglossate archaegastropode redula in Conus striatus, C. marmorus C.flavides and C. lividus. The significance of positive staining

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of redular chitin by the PAS reaction was discussed (Marsh, 1978). Histological description of the different structures of the pharyngeal bulb in the snail Ampularia canaliculata were emphasized. Both morphological and secretary characteristic changes according to the zonal function in relation to development of masticatory structures are worked out. (Catalan et al 1981). The taxanomic value of redula morphology in prosobranch systemics as advanced by Troshel (1886) were confirmed and discussed (Bandel, 1979). Taxonomic revision with secondary sexual characters found in dimorphic Drupella with the use of scanning electron microscopy. Here the rechidian teeth of male are larger than female. (Fujoka, 1982). Buccal mass of the snail Theba pisama was studied by using several histological and histochemical techniques (Roldan and Cosin, 1979).

From the critical analysis of these investigation it is seems that no study is done especially with Cypraean spp. related to histology and histochemistry of mucins in buccal mass. The present investigation tries to present the histological picture and the cellular site of elaboration of the mucosubstances by buccal mass. In the histological observations 4 layered pattern is seen which consist of serosa, muscular layer, submucosa and mucosa. Complex organiazation and occupation of more space on dossal region of buccal mass by muscler

layer must be related to easy and smoth functioning of the redula.

The chitinous material present in redula contains glycogen, neutral mucins and weakly sulfated mucosubstances. Mucosa is mainly formed by columnar cells where goblet cells are occuping major place. These cells can be hemologus, with mucocytes fefered by Hyman (1967). Columnar cells contain in them glycogen and nonsulfated mucins while the goblets are representing glycogen, netural and weakly as well strongly sulfated acidic mucins. Submucosa contains glycogen neutral mucins and weakly sulfated acidic mucins. Mainlyg glycogen and neutral mucins are confirmed in muscular layer and serosa. Differential digestive capacity alloted to the columnar and goblet cells as they contain different types of mucins.

Presence of glycogen in general in all four layers and their cellular componants can be understood as readymade source of energy. Neutral and acidic mucosubstances may be have a role to nulify the posible toxicity produced by the food that has engulfed by the snail. Both sessile, fauna and flora on the surface of the rock and algal material, scrapped as the food by the snail. Another possible role of these substances may be used for digestion of the food to some extent and the protection of the cellular componants of the mucosa layer. Lubricating the redulary appartus and clumping of food particals may be another function to the secretions of

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the mucosa layer.

Estimation of the secretions from the individual cells and their biochemistry may focus light substaisially to understand digestive phenomenon in the lumen of buccal mass.

PRE- OESOPHAGUS

The investigation regarding the mucosubstances in the different histological organization of pre-oesophagus was thought with a view to agument the understanding in general the localization histochemical nature and possible role in physiology of digestion of the marine snail <u>Cypraea arebica arebica</u>.

The lining of oesophagus frequently with short to tall ciliated columnar epithelium. It consist of mucocytes supported inwordly by connective tissue and musche fibres. (Hyman, 1967). In <u>Tonna</u> the epithellial layer is liberally providedw with mucocytes (Weber, 1927). Carbohydrate digestion in the snail <u>Tegula funbratis</u> has been studied and the enzymes laminarase and alginase are reported. Some carbohydrate hydrolysis appears to be due to the bacterial action (Berrie <u>et al.</u>, 1965). Presence of amylase was detected in foregut of <u>cellana rediata</u> (Rao, 1975).

The present investigation dealing with morphology doubts about the presence of salivary gland in the snail.

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However diffused glandular mass without any definate shape has been localized along the dorsal margin of the pre oesophagus. The nature of its histochemistry is not considered since the investigation has been limited to digestive tract only.

The tube of pre-oesophagus comprises four layers serosa, muscular layer, submucosa and mucosa. Mucosa consist of two kinds of cells goblets and columnar. Goblets contain in them glycogen, strongly and weakly acidic sulfomucins and neutral mucins while glycogen, neutral mucins, weakly acidic mucosubstances and sialic acid in them. Observation of glycogen, carboxylic containing sialic acid and strongly sulfated acidic mucosubstances are confirmed in submucosa while glycogen and weakly acidic mucins are stressed in muscular layer. Serosa exhibits glycogen, strongly sufated acidic mucins and carboxylic containing acidic mucosubstances.

Presence of glycogen in the cells of pre oesophagus solves the problem of readymade energy supply. Sulfated acidic mucins and nonsulfated acidic mucins may be lubricating the passage to easy transport of semidigested food to the further part of the alimentary tract. Whether this part of digestive tract has any significance with physiology of digestion is problemetic but mear passage of food smoothly is the definate role that can be alloted to it. Upturn of the tube

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may prevent the food passage back into the buccal cavity. Any other fuctional aspect will be the subject of investigation.

OESOPHAGEAL BULB

Oesophageal bulb as the name suggest represents the expansion of oesophagus like the crop of the avian digestive system. The specilization of this organ in the system as far as its morphology, histology, and histochemistry is concerned must have some definite significance. To get the knowledge of it both histological and histochemical studies are undertaken. It will be helpful to understand the functional role to some extent.

Sometimes the oesophagus at its beginning presents an expansion variously called pyrifom organ, crop, jabot, pharynx of Leiblein and esophageal bulb. The interior of bulb is more or less folded almost transverse in its orientation. Clothing of their folds is with a columnar epithelium which are often tall and ciliated in nature. In Cypraea the bulb is posteriorly located (Hyman, 1967). In <u>Haliotis</u> and <u>Patella</u> the oesophageal bulb has highly folded wall (Graham, 1932). Presence of laminarse and alginase is reported in <u>Tegula funebralis</u> (Berrie <u>et al</u>;1965) Amylase was detected in foregut of <u>Gellans rediats</u> (Rao, 1975). A carnivores mesogastropod <u>Polinieces</u>

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<u>lewsis</u> prey upon bivalve. The anterior region of the gland secretes masked precursor of trypsin activated proteolytic emzyme. It is used for extraorganismic digestion of the prey. The reminder of the gland contain alpha-amylase, alpha-glucosidase, esterases etc. The gland may be absorptive and secretory. (Reid and Friesen 1980). In <u>Tonna</u> the oesophageal caecum is considered to represent the bulb (Weber, 1927).

The investigation carried out in this laboratory reveals the presence of glycogen, neutral mucin and sulfated mucing in goblets and presence of glycogen neutral mucins and sulfated acidic mucosubstances in coloumnar and granular cells of the mucosa layer. Submucosa contain in it glygogen and strong as well as weak sulfomucins in general but the mast cells in them represent glycogen and sulfated acidic mucosubstances. Muscular layer demonstrates glycogen sulfomucins and neutral mucosubstances and both serosa along with peripheral cells contain glycogen and AF positive mucosubstances.

Presence of glycogen in to all histological componants of the oesophageal bulb fulfils the need of energy supply while remaining mucins that are detected must have other physiological significance. Extracelular digestion by producing proteinases may have the important significance because the sac is voluminous and greatly folded which fascinates the secretion of digestive

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enzymes and probably provide place for extra cellular digestion. Presence of glandular cells at the tip of the transvere folds supports the view (plate no.3 fig. no.8). The role of absorbtion of already digested food cannot be over ruled. Morphologically it seems that the oesophageal bulb is modified buccal pouch which lacks in the digestive tract, of this animal which generally occurs in other prosobranches. Elaborated organization and the larger size of the bulb definately awaits a further investigation. Biochemical estimations and enzyme study will sketch a neat alround picture of the oesophageal bulb.

POST OESOPHAGUS

This part of the digestive tract is the posterior containuation of oesophagus after oesophageal gland. Possible role of this tubular part in digestion and the contents of mucins are the viewpoints of the investigation especially the post oesophagus.

Short to tall ciliated columnars epithelium forms the lining of the oesophagus which consist of mucocytes supported inwordly by connective tissue and muscle fibers (Hyman, 1967). In <u>Tonna</u> the epithelial layer is provided with mucocytes (Weber, 1927). The snail <u>Tegula funebralis</u> shows presence of the enzymes laminarase and alginase (Berrie, <u>et al.</u>, 1965). Amylase was detected in foregut of <u>Cellana rediata</u> (Rao, 1975). The extract of83.

the foregut including buccal glands of <u>Tegula Funebralis</u> is strongly amylolytic, hydrolyzing starch, glycogen, sucrose, maltose, laminarin and salicin, but not cellulose, acting best at pH 6 (Galli and Giese 1959).

The present investigation of post oesophagal region is as follows : Goblet and columnar are the kinds of the cells that comprises the mucosa and presence of the sulfated acidic mucosubstances, glycogen and neutral mucins in goblets and glycogen, sulfated mucins and sialic mucins are demonstrated in columnar cells. Submucosaare characterized by the presence of glycogen, neutral mucins and strongly acidic sulfomucins. Glycogen weakly sulfated acidic mucosubstances and sialic acid are confirmed in muscular layer and serosa.

Glycogen must be the available source of energy for the different layers and their cellular organization. Other sulfated and non-sulfated mucins presence may be related to lubrication of the internal canal which may allow the food to pass smoothly. The pessible function of food digestion by post oesophagus seems to be nonexplainable. Increased thickness of the wall by elaboration of muscular layer may possibl associated with perstalsis like movement of post oesophagus.

STOMACH

External delimitation of stomach from post oesophagus is very difficult but it forms the major organ of digestive tract in all animals. Hence the importance is anti-

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cipated and the study of mucins in the histological structures of this organ are considered for investigation.

Oesophagus course posteriorly and enters into stomach called forestomach or proventriculus has been noticed first by Fischer (1904) in Lottia and also in Patella cellana and related forms by Thiem (1907). Stomach is not muscular in having thin circular and longitudinal coats outside epithelium except in Pila where it is thick circular coat (Prashad, 1925). The stomach is reduced in size and is of simpler construction in carnivorus prosobranches (Hyman1967). In many prosobranches the stomach has an anterior evagination termed style sac that contains a rod known as the crystalline style. According to Young (1930, 1932) it is associated with herbivourus diet. Glandular activity is minimal in the stomach of Atlantidae spp. (Martoja, et al. 1976). Carbohydrate degestion in the snail Tengula funebralis has been studied and the major enzymes amylase, laminarase, and alginase are reported. (Berrie et al 1976). Absence of crystalline style and presence of amylase was detected in foregut of Cellana redita (Rao, 1975). Gasric morphology was considered simple and has given significant role in protein digestion. Alkaline proteinases are low in digestive diverticuli which are important site of intracellular digestion (Reid and Friesen, 1980). Several workers

have emphasised the hydrolysis of cellulose (filter paper) or other complex carbohydrates in the stomach by cellulase from style or elsewhere (Yonge <u>st</u> <u>al</u>.1932)

The investigations carried out in the laboratory gives following results. Mucosa is the innermost lining formed by goblet and columner cells. Goblet contain within them glycogen, and sulfated acidic mucosubstances and columner cells are rich with glycogen, neutral mucins and strongly sulfated acidic mucins. Submucosa containing mast cells show the presence of glycogen and strongly sulfated acidic mucins and stomach glands show glycogen, weakly: - Sulfated acidic mucins and stomach glands show glycogen, weakly sulfated acidic mucosubstances and silic acid. Glycogen weakly, acidic sulfomucins and sialic acid are the componants occure within muscular layer and serosa.

Presence of glycogen in general is associated mucins are energy supply. Sulfated and non sulfated acidic mucins are most probably used for the secretions which are used in extra cellular dogestion. Here the stomach glands should be considered in priority. (Plate no.4 fig. no.8). Function of absorption can be also thought to the mucosa lining. Mast cells seems to secrete the secretion used for increasing the permiability of the mucosa wall as seen in the chordates (Bell <u>et al.</u>, 1972). Nature of the secretion of the mast cells and its role in digestive process in the

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stomach of Cypraea needs further investigation.

INTESTINE

The present investigation on the intestine of the marine snail <u>Cypraea</u> <u>arebica</u> <u>arebica</u> expects the understanding of the histological and histochemical nature. Accordingly different staining techniques are employed.

The intestine proceeds anteriroly from the stomach. In lower prosobranches it may be thrown in to loops. (Haliotos) or coils (Patella) while in higher ones it usually runs a more or less streight course to anus. The intestine is lined with a cuboidal or columnar ciliated epithelium, generaly interspersed with mucocytes or other gland cells. The epithelium is substended by connective tissue often containing some muscle fibers, less often a definate mucle layer is present (Hyman 1967). carbohydrate digestion in the snail Tegula funebralis has been studied and amylase, laminarase, and alginase are reported (Berrie et al; 1965). In the intestinal mucosa of the Buccinum undatum epithelil cells found containing small granules appearing like secretary granules of higher vertebrates, It is proposed that these cells corresponds with B cells of the pancreatic tubules. and presents the site of insuline production (Boguist, 1972) Insuline like substance was documented in the cells of intestinal mucosa (Kazakov, 1978).

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The investigation carried out in this laboratory explains morphological features of intestine showing single coil which is intermingled in midgut gland. Mucosa layer which forms innermost lining shows three kinds of the cells namelygoblet coloumner and granular. Glycogen, neutral mucins and weakly sulfated acidic mucins are demonstrated in goblets, glycogen and neutral mucins in columner cells and glycogen, neutral mucins and weakly sulfated mucins are confirmed in granular cells. Submucosa shows glycogen and weak as well as strongly sulfated mucosubstances. Same componants are also seen in mast cells. Gkycogen, neutral mucins and weakly sulfated mucins are the contents of muscular layer where as serosa contains glycogen and sulfated mucins.

Glycogen localization in all four layers of intestine must be giving nourishment as a energy flow. Other mucins are expected to play an important role in the secretion of the digestive enzymes and absorption of digested food. Thin wall of the intestine may have elasticity to accomodate large amount of food to store and allow the digestion to occure for longer time. Nearly streight course of the intestine with single spire is definately a forword step towords evolution of the alimentary tract. However details of digestion and absorption in this part of digestive tract are yet to be emplored.

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RECTUM

The investigations regarding the mucosubstances in the different histological componants of the rectum are considered with the view of understanding in general the localization, histochemical nature and possible role of physiology of digestion in the marine snail <u>Cypraea</u> <u>arebica arebica</u>.

A widen terminal part of the intestine may be named as rectum. Intestine in the mantle after mmerging from visceral mass is at the roof and ends as anus at the free adge of the mantle. Interstine in mantle either passes through pericardium (Haliotidae, Fissurellidae, Trochidae, Neritidae and Turbinadae.) It may run along the edge of the pericardial cavity. Near the anus ciliary lining of epithelium may be replaced by cutical. (Hyman, 1967). Intestine near the anus is provided with an anal gland in number of the prosobranches and gives dark colour. Such gland is seen in Fissurellidae (Pelseneer, 1898, Speltor, 1928) Muricidae (Fretter, 1946) and Naticidae - Oliva (Kuttler, 1913). The epithelium consist of one kind of cell, ciliated and contains brown granules which are discharged in to rectum for elimination. In the snail Tegula funebralis the enzymes amylase, laminarase and alginase are reported. (Berrie et al., 1976). No enzyme activity was seen in the hindgut of <u>Cellana</u> rediata (Rao 1975). According to Gabe (1951) the anal gland produces mucoid secretion.

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Passage of food through the digestive tract requires 24 to 48 hours (Hirsch, 1925).

All four layers characteristics of the digestive tract are exhibited by the rectum. Presence of the goblets in less papulation contradicts thenature observed in rectum of the chordates where they are profusely papulated. Mucosa layer which is formed by goblets and columnar cells contains glycogen, neutral mucins and weakly sulfated mucosubstances. Submucosa shows the presence of glycogen, and strongly sulfated acidic mucins while mast cells in them are representing glycogen, neutral mucins and strongly sulfated acidic mucins. Glycogen and sulfated mucins are characterized by muscular coat and serosa. Typical brush border is observed in the mucosa wall of rectum and in consists of neutral mucins and strongly sulfated micins.

Glycogen content is the source of the energy for activation of cellular componants. Other mucins are seems to responsible for the lubrication and theeasy passage of the undigested food. Presence of strongly sulfated acidic mucins in the brush border may act on microflora and fauna that accidantly enters in the rectum through mantle cavity or through digestive tract proper. Brown cellular bodies are supposed to be the excretory products that might be thrown into the lumen of the rectum. Absence of the rectal glands and its significance in this animal are unexplainable.