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POST-HATCHING AGE CHANGES OF THE OESOPHAGUS OF TILAPIA FISH (*OREOCHROMIS NILOTICUS*) LIGHT AND TEM STUDIES

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SUMMARY

The development of the oesophagus of 40 oreochromis niloticus fish at different posthatching ages was described using light and TEM. In newly-hatched larvae, the oesophagus appeared as a short tube lying dorsal to the yolk sac. It was initially lined by 3-5 cell layers thick. Three days posthatching, the superficial epithelial layers were expanded in several sheets toward the lumen. At the age of 8 days, the uppermost layer of the epithelium became formed of cuboidal cells with convex free surface. The luminal margin of superficial cells carried prominent microridges. Gradual flattening of these cells occurred till became stratified squamous at 14 days old larvae when the mucosal folds were firstly developed.

At the time of hatching, the mucus cells began to develop inbetween the oesophageal epithelial cells. By the 3rd day, they became filled with vacuolated cytoplasm. At 8 days, the mucus cells began to take the goblet-shape. The mucus droplets were more electron dense in the anterior oesophagus than that of the caudal part. Primordial taste buds appeared as localized groups of undifferentiated cells at 6 days posthatching. Later on, at 8 days, their constituent cells became differentiated into spindle-shaped cells and basal ones. Generally, there was a tendency for taste buds to occur in the anterior part of the oesophagus toward the pharynx. They were gradually decreased caudally till completely disappeared near the stomach whereas the mucus cells were more numerous in the posterior part of the oesophageal epithelium. From 14 days posthatching onwards, the oesophagus attained the four tissue layers' ar-

rangement characteristic of the adult.

INTRODUCTION

The production of fish and fish products in Egypt is considered as a very important source of protein, which is requested nearly by all classes of population. Therefore, it is essential to improve fish quality and quantity. However, fish change their food habits during their life cycle and accordingly suitable changes in the structure of their feeding and digestive organs take place (Nikalsky, 1963 and Ferraris et al., 1987).

Although considerable informations were available on the histology and ultrastructure of the oesophagus of fish. Nevertheless, few studies conducted on the histogenesis of this organ in some fish species were offered (Gustafson and Wolpert, 1967 and Burke, 1981) in sea urchin; (Grizzle and Curd, 1978) in legperch (Ferraris et al., 1987) in milkfish; (Ismail, 1994) in grass carp and karmout; (Bisbal and Bengtson, 1995) in summer flounder; (Otake et al., 1995) in pike eel and (Chia, 1977 and Crawford and Martin, 1998) in star fish. None of the existing literature have been described the same subject in orechromis niloticus (Tilapia) in spite of their commercial importance and their intensive aquaculture interests.

This study, therefore, examines the developmental changes that accompany the posthatching growth of the oesophagus of oreochromis niloticus. This could serve to identify and realize the feeding habitant of this fish and give the appropiate informations to the farm breeder about the optimal time and way of commercial diet use.

MATERIAL AND METHODS

The specimens used in this study were collected from a commercial breader. Larvae were collected daily from 0 day till 14 days then weekly till 4 months after hatching.

For light microscopic examination, 40 fish were used. The whole small sized fish up to the age of one month and the oesophagus samples from the larger sized ones were fixed in neutral buffered formaline and Susa fluids. They were processed and embedded in paraplast. Cross and/or sagital step-serial sections 4-6 um thick were cut and stained with Haematoxylin and Eosin (H & E). Crossmon's trichrome, Gomori's reticulin, periodic acid Schiff (PAS), Alcian blue and Best's carmine (Crossmon, 1937 and Drury and Wallington, 1980).

For TEM, 1.0 mm^3 tissue samples were embeded in 2.5% glutaraldehyde buffered at PH 7.2 - 7.4 with 0.1 M sodium cacodylate, rinsed in the same buffer, and immersed in 1.0% osmium tetroxide in 0.1 M cacodylate at the same PH. The tissues were then dehydrated, cleared in propylene oxide, and embedded in epon resin (Hayat, 1989).

"Thick" survey sections (1.0 to 1.5 um) were cut with a glass knife, and stained with toluidine blue for light microscopy. Ultrathin sections were cut with a diamond knife, stained with uranyl acetate and lead citrate and examined using Jeol TEM 100 CX II at 80 Kv.

RESULTS

In newly hatched larvae, the oesophagus appeared as a short tubular structure lying dorsal to the yolk sac (Fig. 1). It proceeded from the developing pharyngeal cavity to be continued caudally with the developing stomach. The oesophageal lumen was narrow and slightly irregular throughout its length but it became wider and more irregular toward the developing stomach. It was lined by a stratified epithelium of 3-5 cell layers thick. These epithelial cells had ill-distinct cell boundaries, homogenous acidophilic cytoplasm and ovoid or elongated darkly stained nuclei. Some of these cells underwent mitotic division. Mucus cells began to develop inbetween the oesophageal epithelial cells. They appeared as ovoid cells with unstained clear cytoplasm (Figs. 2 & 3). Fine structure of the newly hatched oesophageal epithelium revealed marked changes in the orientation of epithelial cells from the base to the surface. The basal cells were relatively small and a large part of their volume was occupied by the nucleus. The sparse cytoplasm of these cells was rich in free ribosomes, mitochondria and scant rER (Fig. 3). The cells immediately above the basal layer showed some striking changes represented by the great development of rER, electron lucent cytoplasm, Golgi apparatus and cytoplasmic vacuoles. The superficial cells carried short microridges and revealed relatively small nucleus comparing with the basal ones. The cytoplasm of these cells was filled with free ribosomes, few rER and few dense granules. Intercellular spaces appeared inbetween (Fig. 3).

The underlying propria submucosa was formed of a mass of undifferentiated mesenchymal cells embedded in an amorphous intercellular substance. This layer was encircled by a relatively thin layer of developing muscle cells that was differentiated from the mesenchymal cells. The latter layer was followed by the developing tunica adventitia. It was formed of highly cellular vascularized mesenchyme (Figs. 1 & 2).

Three days after hatching, the superficial layers of the lining epithelium were expanded in several sheets which projected into the lumen (Fig. 4). Desquamation of some of the inner cells was noticed. At the same stage of growth, the mucus cells were greatly increased in number and size. They became filled with many mucus droplets that pushed the nucleus toward the base of the cell (Figs. 4 & 5). They appeared irregular or ovoid darkly stained and surrounded by a thin rim of cytoplasm. This lining epithelium was abruptly changed into the columnar epithelium of the developing gastric mucosa (Fig. 4).

By TEM, the basal epithelial cells rested on a thin basement membrane appeared to have electron dense nuclei and cytoplasm (Fig. 5). The latter contained mitochondria and abundant ribosomes. The adjacent cells showed obvious intercellular spaces and the lateral membranes were joined together by desmosomes. The main bulk of the midway between the basal and superficial cells was occupied by undifferentiated cells and leukocytes (Fig. 5). The cytoplasm of these cells was characterized by an electron dense matrix, rER, mitochondria and free ribosomes. The surface epithelium showed mucus cells and less electron dense cells. The latter carried microridges. At the same time, further growth and development of the muscle fibers took place. As a result, the developing tunica musculosa became thicker and appeared to be formed of one layer of circularly arranged muscle fibers (Fig. 4).

By 6 days posthatching, the oesophageal tube was markedly increased in both length and thickness (Fig. 6). Toward the pharynx, the epithelial folds became fewer and lower, while toward the stomach they appeared more numerous and elongated and occasionally, they might obliterate the lumen. The desquamation of the inner epithelial cell sheets was greatly decreased. However, the mucus cells became more numerous in the posterior part of the oesophagus than in the anterior one (Fig. 6). At the same time, primordial taste buds could be detected for the first time. They appeared as localized groups of undifferentiated cells scattered among the anterior part of the oesophageal epithelium (Fig. 6). Differentiation of the underlying mesenchymal cells into fibroblasts began to appear in the developing propria-submucosa. Moreover, inner longitudinal muscle fibers start to develop. Some of these muscle fibers extended into the submucosa.

At the age of 8 days posthatching, absorption of the yolk material was begun. The developing taste buds became easily distinguishable from the surrounding epithelial cells (Fig. 7). Their constituent cells became differentiated into spindle-shaped cells and basal ones. The latter cells possessed spherical or ovoid nuclei and acidophilic cytoplasm. The spindle-shaped cells had oval or elongated nuclei and acidophilic cytoplasm. Some of these cells had pale vacuolated cytoplasm. However, the apical surface of the taste buds was covered by the uppermost superficial layer of the surface epithelium. The latter layer was made up of cuboidal cells with convex free surface, many of which were dome-shaped. They had dense acidophilic cytoplasm. Their spherical or ovoid shaped nuclei were moderately stained and oriented parallel to the surface of the oesophageal mucosa. These cells also covered the apical surface of some developing mucus cells (Fig. 8). Some of the latter cells took the goblet shape. They acquired a narrow neck that extended deeply to the area immediately above the basal cells of the oesophageal epithelium. However, the apical surfaces of some mucus cells might open into the lumen by small pores (Figs. 8 & 9).

Tight junction was clear between the superficial cells and the apical part of mucus cells. Moreover, some of the mucus droplets were enclosed within a limiting membrane while many of these droplets were fused with each other (Fig. 9). At the mean time, the oesophageal muscles were progressively increased in thickness. However, the inner longitudinal muscle fibers were present in some areas but absent in others (Fig. 7). Such muscle fibers became multinucleated and had acidophilic cytoplasm that showed fine cross striations. Moreover, some of these muscle fibers extended obliquely into the submucosa (Fig. 8).

By the 11th day posthatching, taste buds and mucus cells appeared more numerous in the oesophageal epithelium. The latter rested on an undulated reticular basal lamina (Fig. 10). Fine reticular fibers were also demonstrated in the propria-submucosa.

As the oesophageal development advanced particularly from 14 days posthatching till the adult stage, the surface epithelium was gradually increased in thickness and showed progressive flattening of the superficial cells. The oesophageal mucosa began to project into many mucosal folds. The cores of these folds were filled with the connective tissue of the propria- submucosa (Fig. 11). The taste buds became oval elongated in shape and occupied nearly the entire thickness of the epithelium. They might be communicated with the connective tissue papillae projected from the underlying lamina propria (Figs. 11 & 12). The constituent cells of the taste buds appeared more differentiated. The spindle-shaped cells became thinner and acquired PAS-positive tapering ends with hair-like terminals which might reach the taste pore (Fig. 12). Their nuclei were elongated and darkly stained. The contents of mucus cells were intensely reacted to PAS (Fig. 12), alcian blue (Fig. 13) and toluidine blue (Fig. 14), while they gave a negative reaction with Best's carmine. Near the stomach, the stratified oesophageal epithelium was characterized by abundant mucus cells. This was replaced by a single layer of gastric columnar cells followed by alcian blue-positive gastric glands (Fig. 13).

In adult fish, there was a progressive increase in the number and size of the mucosal folds. There was a tendency for mucus cells to occur in the posterior part of the oesophagus toward the stomach (Fig. 14). On the other hand, the taste buds were more numerous in the anterior part toward the pharynx (Fig. 15). Mucus cells contained numerous mucus droplets; most of them were enclosed in a limiting membrane (Figs. 16-18). The thin lateral cytoplasm of such mucus cell had less electron dense ribosomes, few mitochondria, rER and the rest of the cytoplasm was filled with many mucus droplets and contained basally situated nuclei (Fig. 16). The mucus droplets were more electron dense in the anterior oesophagus (Fig. 17) than that of the caudal part (Fig. 16).

The superficial cells had electron lucent irregular nuclei. The cytoplasm contained few minute mitochondria, free ribosomes, few rER and electron dense granules. Granules had the same density like that of cytoplasmic ones were noticed in the lumen. These luminal cells revealed large vacuoles, which increased in the protruded part of the cell (Fig. 17). Well developed Golgi complex was extensively apparent (Fig. 18 & 19). The plasmalemma of the adjacent cells interdigitated extensively (Fig. 17) whilst the lateral membranes facing the mucus cells presented complicated tubular indentations and intricate contact (Fig. 18). The luminal margin of each cell was characterized by the peripheral microridges (Figs. 16-19). The pattern of microridges born by each cell was distinctive of that cell. In general the arrangement of microridges was such that a number of contiguous segment ran parallel to each other forming a unit, which gradually blended with adjacent units where the microridges were oriented differently. The microridges were almost covered by mucus coat (glycocalyx) (Fig. 19).

The muscular coat showed complete differentiation in muscle cell type and arrangement. The muscle fibers were progressively increased in size and the cross-striation became very obvious (Figs. 14 & 15). These muscle fibers were arranged in two distinct layers, inner longitudinal and outer circular. The tunica adventitia of the anterior part of the oesophagus and the serosa of the posterior part showed a relative increase in the thickness and amount of connective tissue.

DISCUSSION

In this manuscript, the oesophagus of newly hatched larvae is lined by stratified epithelium of 3-5 cell layers thick. By the 3rd day posthatching, the superficial cells expand in several sheets toward the lumen. This was true in perch (Hirji, 1983) and pike eel (Otake et al., 1995). At the early larval stage in our work, the epithelium is formed of basal columnar cell that showed gradual flattening with advantage age and become squamous in the surface. Bisbal and Bengtson (1995) recorded the same result in summer flounder. The cytoplasm of these basal cells contain free ribosomes, mitochondria and scant rER. in Tilapia oesophagus. The cytological features of these cells seemed to carry the same criteria of undifferentiated mother cells found in Luderick (Anderson, 1986). These cells were progressively proliferated and the generated cells move upward toward the surface in perch (Hirji, 1983). Stratification of the oesophageal epithelium served to provide thick renewable protective lining (Martin and Blaber, 1984).

Another undifferentiated light cells with vacuolated cytoplasm are noticed immediately above the basal epithelial cells. These cells accumulate free ribosomes, Golgi apparatus and rER. in electron lucent cytoplasm. Such cells could be accepted as the forerunner of the future mucus cell (Hirji, 1983).

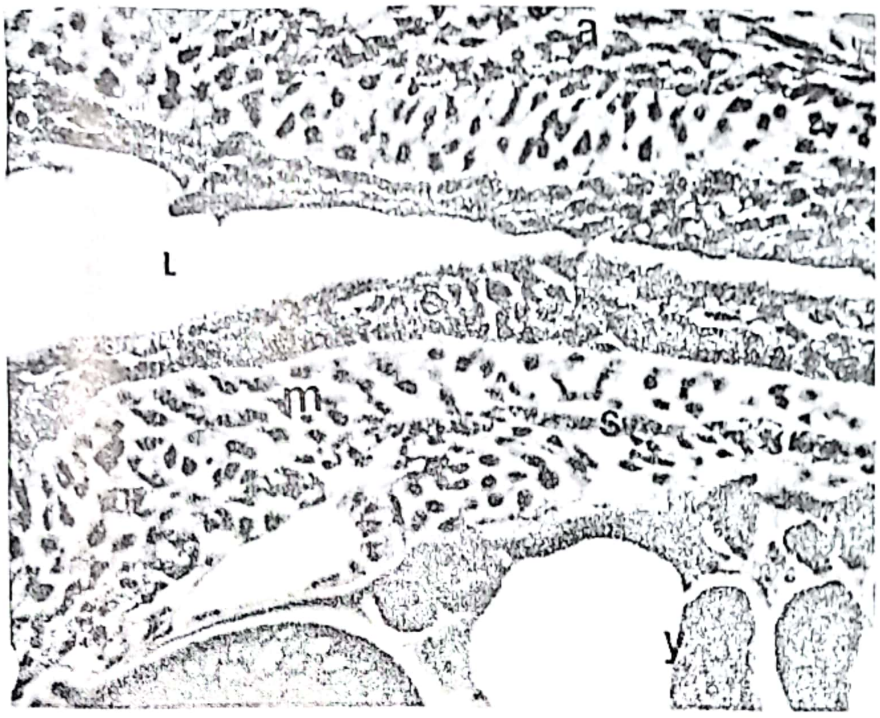

Fig. (1): Longitudinal section through one day posthatching fish showing the oesophageal epithelium (e), mesenchymal tissue (m), muscle layer (s) and tunica adventitia (a). Notice that the oesophageal lumen (L) became wider toward the developing stomach. Notice also the yolk sac (y). H & E stain, X 410.

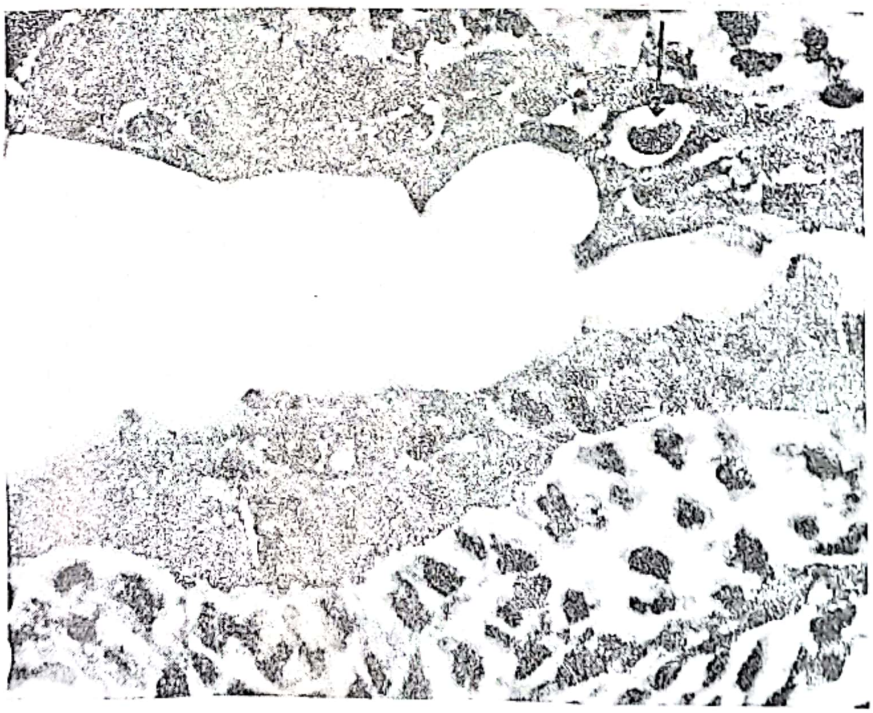

Fig. (2): A high magnification of Fig. 1 showing the appearance of mucus cells (arrows) in between the oesophageal epithelial cells. H & E stain, X 800.

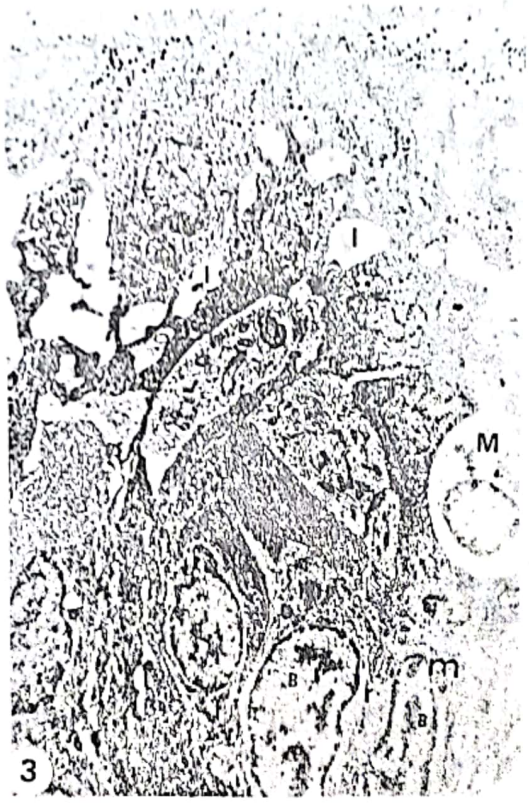

Fig. (3): Electron micrograph of the oesophagus of one day larva showing the basal cell (B) contain scant r.E.R (r) and mitochondria (m). Notice the forerunner of mucus cells (M) and the intercellular space (I). Uranyl acetate - Lead citrate X 6500.

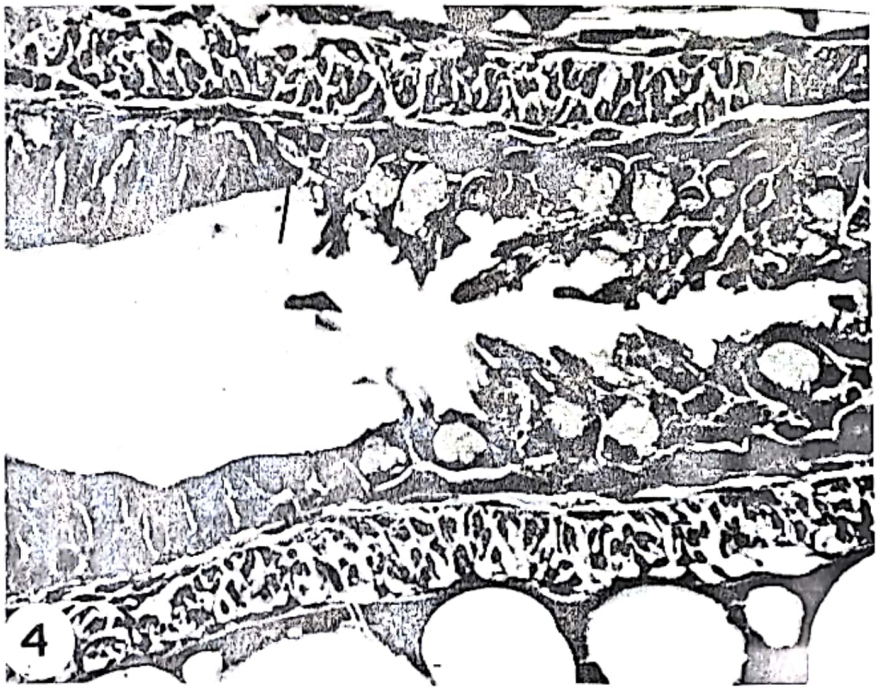

Fig. (4): Longitudinal section throu gh 3 days posthatching fish showing the extension of the oesophageal epi. ielium into the lumen. The mucus cells became filled with vacuolated cytop asm. Notice the abrupt change of the oesophageal epithelium into the colu nnar epithelium of the gastric mucosa (arrow). H & E stain, X 410.

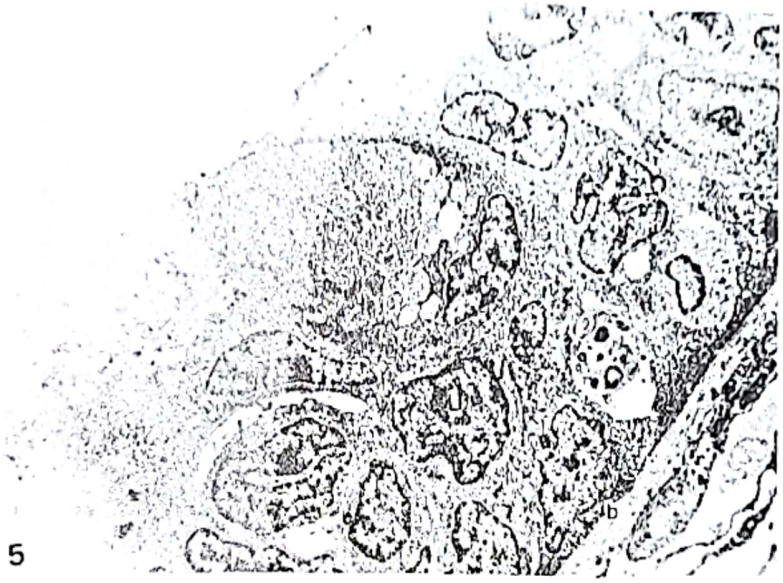

Fig. (5): Electron micrograph of the oesophagus of 3 days larva showed the basal cell (B) had electron dense cytoplasm and rest on a thin basement membrane (b). Note the undifferentiated cells (U) and Leukocytes (L). Uranyl acetate - Lead citrate X 6500.

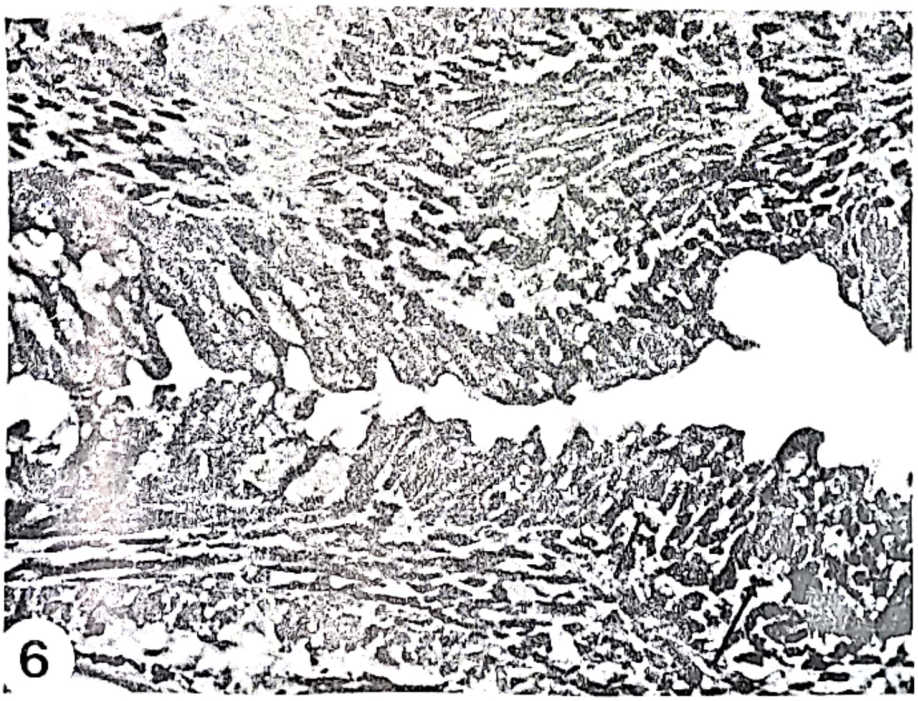

Fig. (6): Longitudinal section through 6 days posthatching fish to show numerous mucus cells in the posterior part of the oesophagus and less so in the anterior one. Notice the first appearance of the taste buds (arrows). H & E stain, X 320.

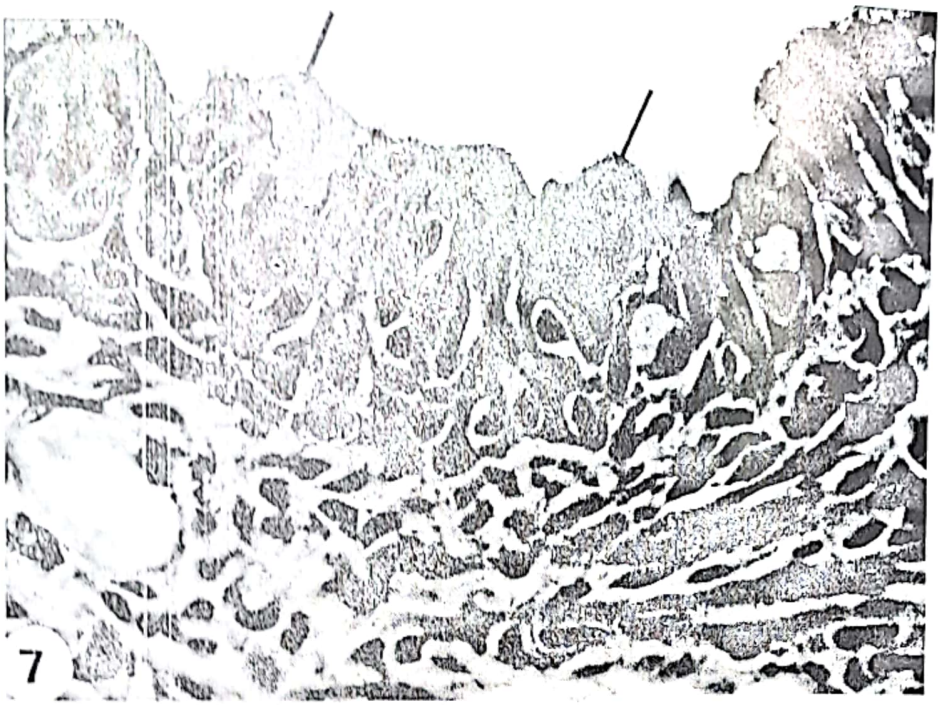

Fig. (7): Anterior part of the oesophagus of 8 days posthatching fish showing dome-shaped cells (arrows) in the uppermost layer of the oesophageal epithelium. Inner longitudinal muscle fibers, with fine cross striations, started to appear in some areas. Notice the spindle- shaped cells and the basal ones of the taste buds. H & E stain, X 800.

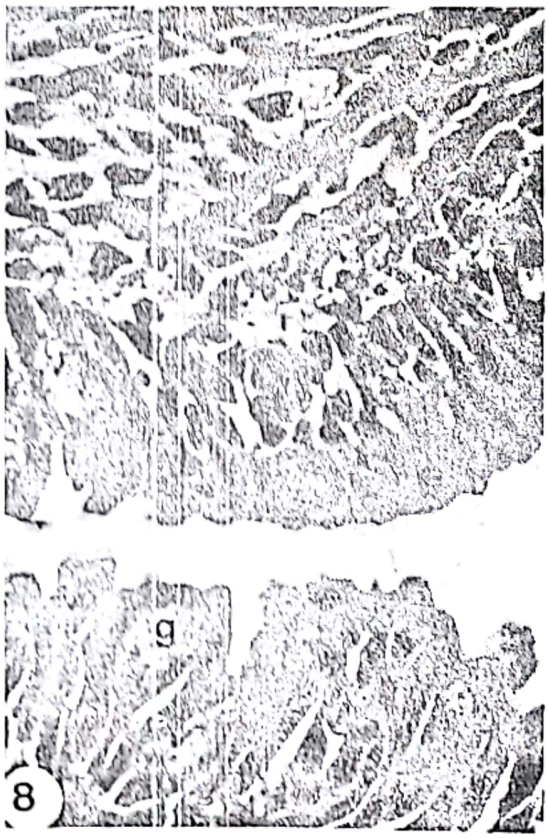

Fig. (8): Posterior part of oesophagus of 8 days posthatching fish to show that some of the mucus cells took the goblet-shape (g). Notice that some of the muscle fibers extended obliquely into the submucosa. H&E stain X 800.

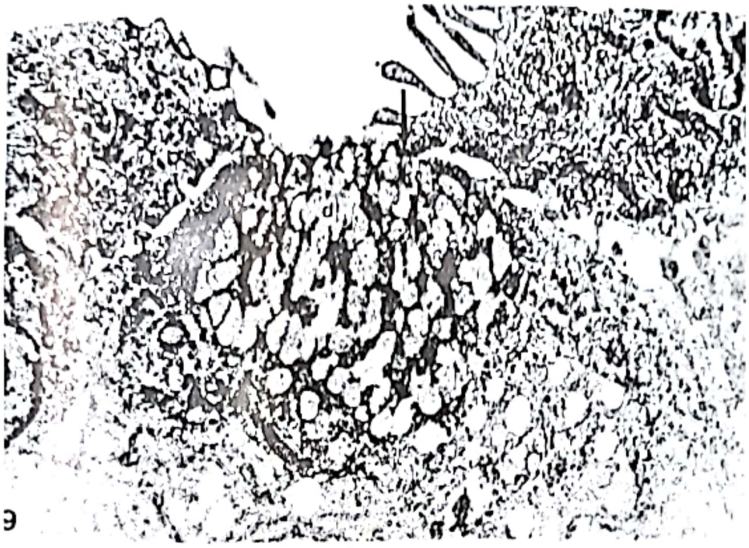

Fig. (9): Electron micrograph of the oesophagus of 8 days larva showed tight junction (arrow) between the superficial cells (S) and mucus ones. Notice the fusion of mucus droplets (d). Uranyl acetate - Lead citrate X 14,000.

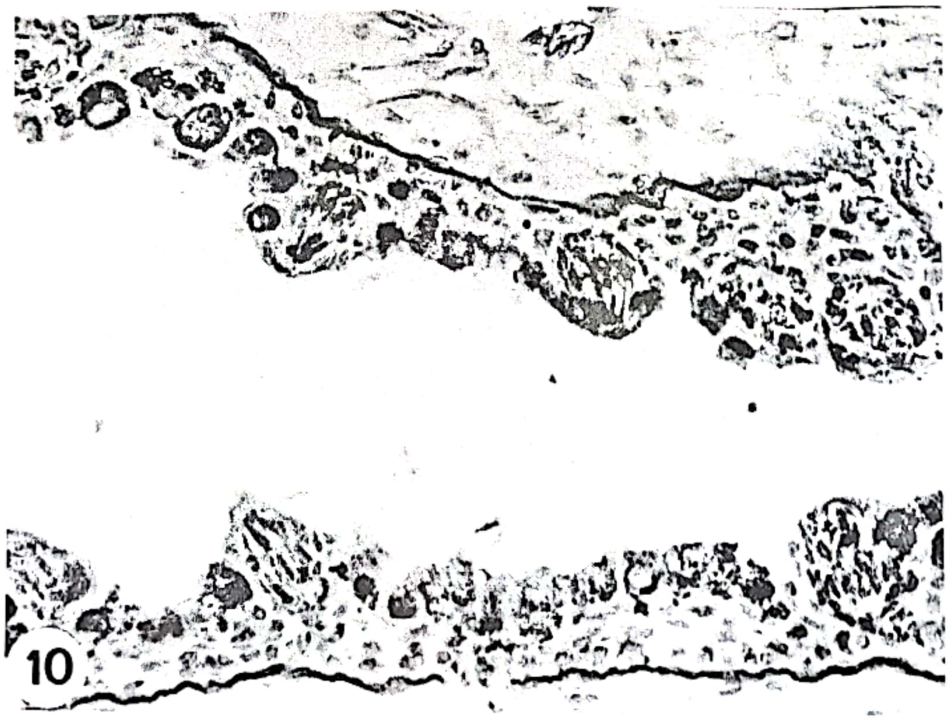

Fig. (10): Growing oesophagus of 11 days posthatching fish showing the subepithelial condensation of reticular fibers. Gomori's reticulin method, X 410.

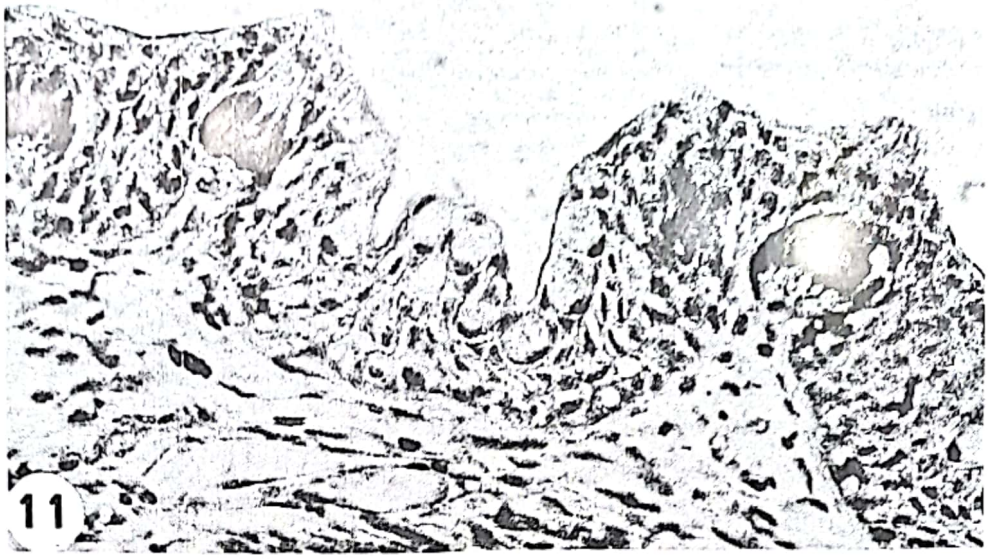

Fig. (11): Oesophageal mucosa of 14 days posthatching fish showing the beginning of formation of mucosal folds. Notice that the taste buds became communicated with the connective tissue papillae projected from the lamina propria. Crossmon's trichrome stain, X 410.

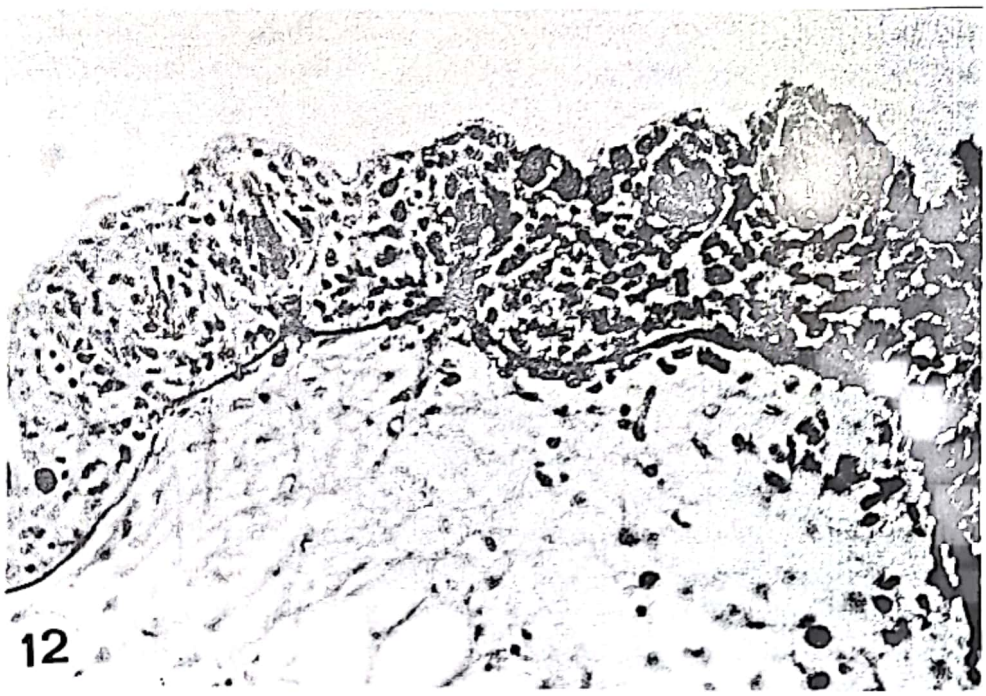

Fig. (12): Anterior part of oesophagus at 20 days posthatching fish showing PAS reactivity in the basement membrane, some of the taste buds cells and taste hair. Notice the strong PAS-positive material in the mucus cells. PAS technique, X 320.

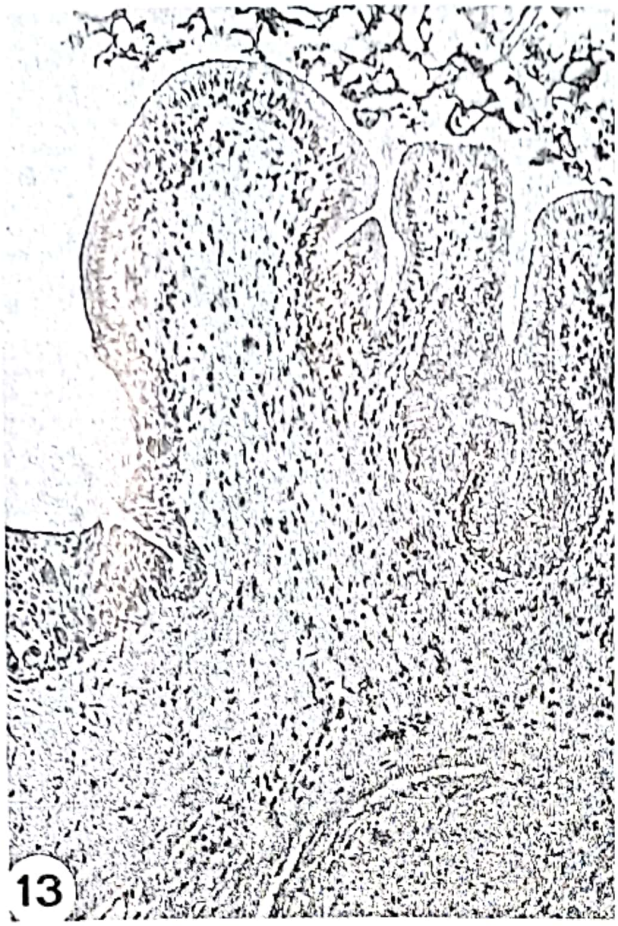

Fig. (13): Gastro-oesophageal junction at 24 days posthatching fish showing alcian blue reactivity in the mucus cells and gastric glands. Alcian blue stain. X 130.

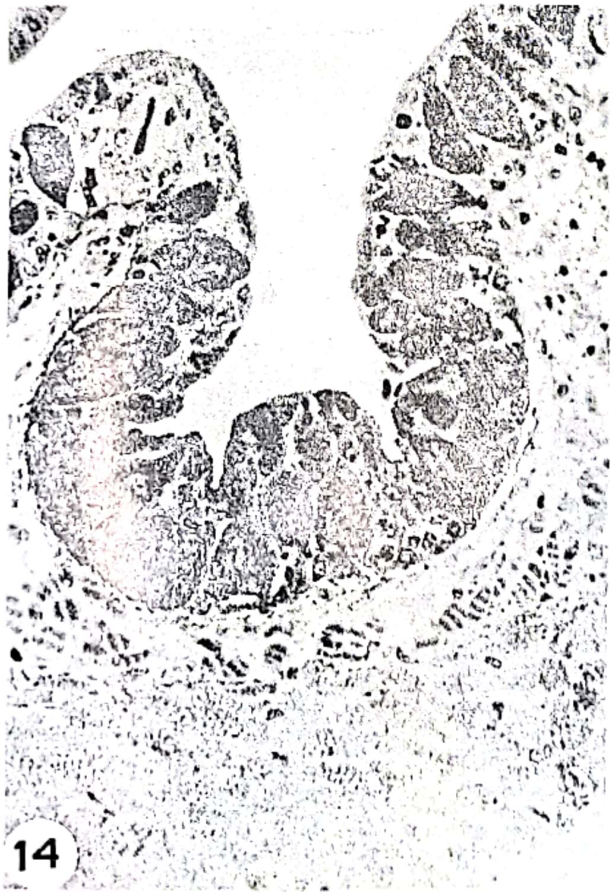

Fig. (14): A lot of mucus cells located in the posterior part of oesophagus of adult fish. Toluidine blue stain, X 320.

Fig. (15): Longitudinal section through the anterior part of oesophagus of adult fish showing completely developed taste buds. Notice the multinuclear character and the clear cross striations of the inner longitudinal muscle fibers of the tunica musculosa. H & E stain, X 800.

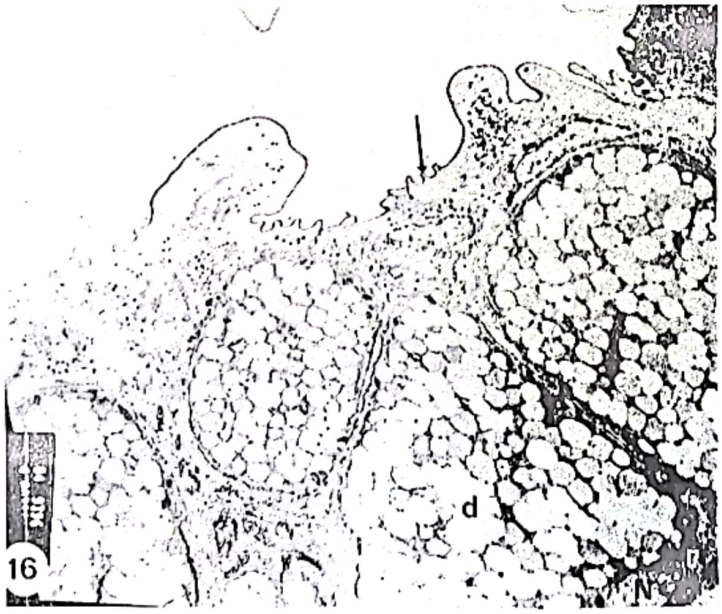

Fig. (16): Electron micrograph of the posterior oesophagus of adult fish showing peripheral microridges (arrow). The mucus cell had few mitochondria (m), r.E.R. (r), basal nucleus (N) and electron lucent mucus droplets (d). Uranyl acetate - Lead citrate X 2000.

Fig. (17): Electron micrograph of the anterior oesophagus of adult fish illustrated the superficial cells (S) which revealed few mitochondria (m), few rER. (r), vacuoles (v) and electron dense granules which found also in the lumen (arrows). Notice the interdigitation (n), microridges (arrow head) and electron dense mucus droplet (m). Note also the protruded part of the cell. Uranyl acetate - Lead citrate X 10,500.

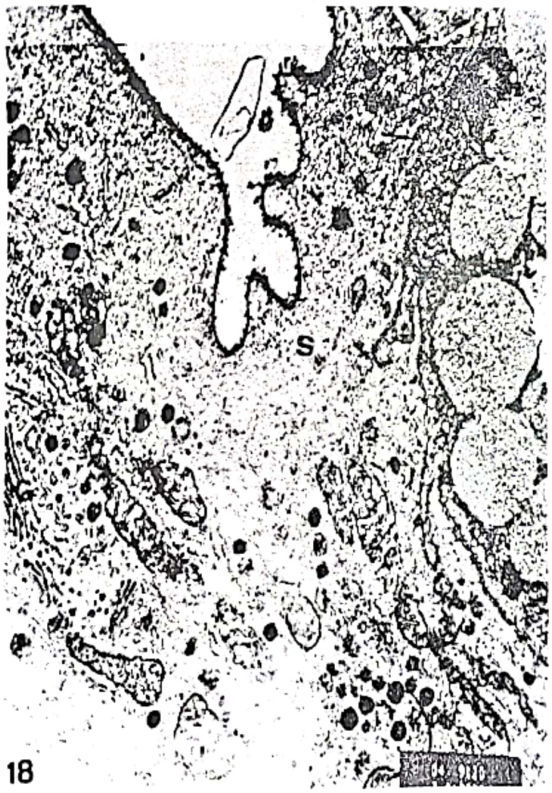

Fig. (18): Electron micrograph of the oesophagus of adult fish demonstrated the superficial cells (S) contained well developed Golgi apparatus (G). Notice the tubular indentations between the superficial cell and mucus cell (arrow). Note also the microridges (r) and the mucus droplets enclosed in limiting membrane (arrow head). Uranyl acetate - Lead citrate X 17,000.

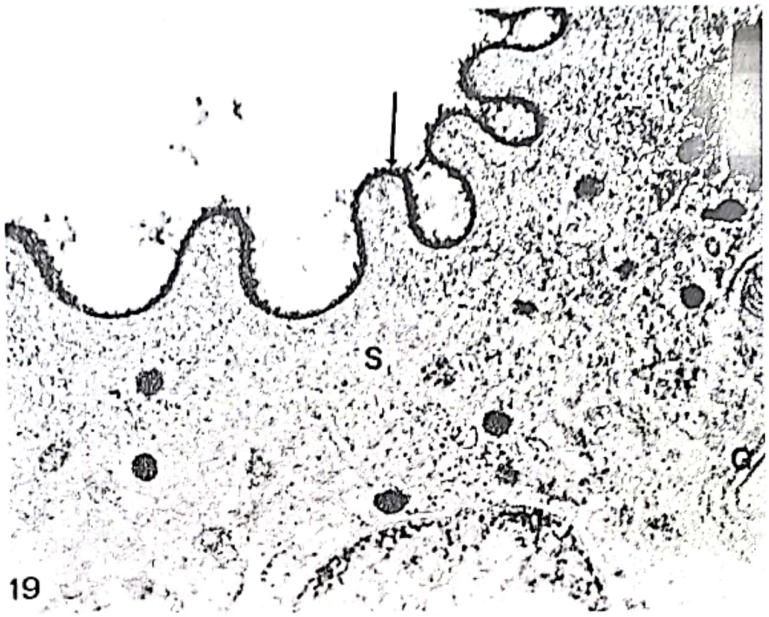

Fig. (19): Electron micrograph of the oesophagus of adult fish revealed the Golgi apparatus (G) of the superficial cells (S). Note the clear microridges that covered by a mucus layer (arrow). Uranyle acetat - Lead citrate X28,000.

As the age proceeded, the stratification increases in thickness and the surface epithelium is occupied by two prominent cell types, mucus cells and superficial cells. The mucus cells noticed in this work have been described by Al-Hussaini (1947) in Atherina forskall, Islam (1951) and Al-Hussaini and Kholy (1953) in teleost, Kapoor (1958) in Gadusia Chapra, Pasha (1964) in Tilapia mossambica, Reifel and Travil (1977) in telostean, Hirji (1983) in perch, Anderson (1986) in Luderick (Ismail, 1994) in grass carp and Karmout, El-Shammaa et al. (1995) in karmout and Mokhtar (1995) in marine fish.

Our histochemical investigation is great similar to that of Reifel and Travil (1977) that mucus produced by these cells is stained positive with PAS and alcian blue. This could indicate that the mucus secretion contains both acid and neutral mucopolysaccharides (Sis et al., 1979 & Yoakim and Khidr, 1985) in catfish (Cataldi et al., 1993) in mugil cephalus, (Ismail, 1994) in grass carp and Karmout and (Mokhtar, 1995) in marine fish. The chemical composition of mucus may explain that the mucus serve as a lubricant and probably enable the oesophegus to play its primary role as a transit tube for food from the oropharyngeal cavity to the stomach (Ezeasor and Stokoe, 1980) and our study. The mucus also may act in iono-regulation as a charged layer (Marshall, 1978 and Simonneaux et al., 1987) in teleosts, modifying ion concentration at the cell contact (Kirschner, 1977) in anguille and could be an important component in the osmoregulatory function of the gut particularly the oesophagus (Kirsch, 1978 and Kirsch et al., 1985) in teleosts.

The present study shows that the mucus coating differs between the anterior and posterior oesophagus, representing dense globules within the anterior part and electron lucent in the posterior part. This extreme in density in fact could support the hypothesis of Humbert et al. (1984) that the dense mucus layer of the anterior part may be protective and essentially involved in removing excess of water from food particles during swallowing. Whereas, the electron lucent mucus may be involved with absorption and be a site of selective ionic diffusion.

The superficial cells have a few mitochondria, free ribosomes, few rER., dense granules and many vacuoles. These dramatic changes may support the idea of (Hirji, 1983 and Ismail, 1994) in grass carp that the appearance of many vacuoles in the superficial cells represents a sign of aging and the cells are shed. Hirji (1983) added that the underlying migrating cells replace them. The physiological function of these cells is not known. Physically they may provide mechanical support to the pores of mucus cells in pike (Linss, 1969) and also can protect the rest of mucosa in teleost (Reifel and Travill, 1977) and in Mugil cephalus (Cataldi et al., 1993).

The presence of dense granules in the cytoplasm of the superficial cells was discussionable.

These granules were thought to be digestive enzymes in Esox lucius (Linss and Geyer, 1968). However, they might be involved in vacuolation process taking place in these cells (Reifel and Travil, 1977). In oreochromis niloticus the dual functions could be accepted. However, the appearance of these dense granules evolved in the oesophageal lumen probably confirmed that they may be digestive enzymes and digestion could start in the first region of digestive canal (Sabapathy and Teo, 1993) in Siganus. Also, vacuolation of these cells in our study may support the second hypothesis.

The primordia of taste buds is noticed by 6 days of development and become easily detected by the 8th day posthatching. Thereafter, at 14 days the taste buds increase in size and number and occupied mostly the anterior part of oesophagus near to the pharynx. At this particular time, the larvae began to expose to external feeding as the yolk sac started to disappear (Ferraris et. al.; 1987, and El-Habback, 1995). The presence of taste buds in teleosts was previously reported by several investigators in teleosts (Mohsin, 1962 and Reifel and Travil, 1977), in Oryzias Latipes (Hamed et al., 1984), in cod (Bishop and Odense, 1966), in Trout (Ezeasor and Stoke, 1980) in grass carp and Karmout (Ismail, 1994) and in marine fish (Mokhtar, 1995). Taste buds are essentially chemoreceptors, and accordingly, the anterior oesophagus could be important for more specific selection of food before swallowing (Bishop and Odense, 1966; Ezeasor and Stoke, 1980 and Hamed et al., 1984).

The major feature of the oesophagus is the sculpting of luminal plasmalemma of the superficial cells into microridges. Weinreb and Bilstad (1955) in rainbow trout, Sperry and Wassersug (1976) in cat fish and Humbert et al. (1984) in Anguilla confirmed the presence of these ridges in the oesophagus of fish. Since microridges have been shown to exist on epithelial surfaces such as skin and cornea which are subjected to mechanical insult (Harding, 1973). The microridges therefore may represent a mechanical adaption which, in the oesophagus; would withstand the trauma resulting from ingested materials (Sperry and Wassersug, 1976). In addition, the microridges would serve and hold a film of mucus secreted by mucus cells (Ezeasor ans Stoke, 1980) in trout. Microridges have been specifically implicated in a mucus-retention mechanisms (Olson and fromm, 1973; Hawkes, 1974; Andrews, 1975 and Schliwa, 1975) and our observations are consistent with that interpretations. Sperry and Wassersug (1976) added that microridges insured that as the food passes in the oesophagus, it does so on a continuous coat of mucus material.

In the present study, the mucosa of the oesophagus shows the first appearance of folds at 14 days posthatching. However, the development of these folds at early life stage of Tilapia fish may come at the time showing change in fish habitat. Longitudinal folds along the whole oesophageal

length are detected in advanced age. This finding was true in sea bass (Blake, 1930), Carp (Curry, 1939), Mulloides auriflamma (Al-Hussaini, 1946), Trout (Anderson and Mitchum, 1974), Common eel (Clarke and Witcomb, 1980), striped bass (Groman, 1982), Salmonides (Yasutake and Wales, 1983), milkfish (Ferraris et al., 1987) and Karmout (El-Shammaa et al., 1995). These folds could provide large surface area for the passage of food in the oesophegus (Clark and Witcomb, 1980). Dispensability of the oesophagus is permitted by the presence of longitudinal folds (Reifel & Travill, 1977). Moreover, El-Shammaa et al. (1995) claimed that these folds could break down the ingested bolus into smaller portions for efficient mixing with the gastric fluid in the stomach.

At the day of hatching, the future tunica muscularis is formed of thin layer of developing muscle cells. Three days later, thick circular layer is formed which acquired inner longitudinal layer by the 6th day of development. Multinucleated striated cells appear by the 8th day of development which increase gradually in size to form inner longitudinal and outer circular skeletal muscle in adult. Similar studies have demonstrated the same development sequence of the oesophageal muscle cells, (Gustafson and Wolpert, 1967 and Burke, 1981) in sea urchin and (Chia, 1977 and Crawford and Martin, 1998) in Starfish. The presence of oesophegeal circular muscle may imply that the oesophagus has a mixing or triturating effect on the food (Anderson, 1986; Ismail, 1994 and Mokhtar, 1995). This explanation is in the line with the hypothesis that the oesophageal muscle must breakdown the cell walls of ingesta in order to prepare it for digestion and help in passing food from mouth to stomach (Crawford and Martin, 1998). The longitudinal layer may serve as reinforcement to oesophagi subjected to violent extension by ingestion of food (Yoakim, 1968).

In our study, the close timetable of development and coexistence of taste buds and striated muscles in the anterior esophagus reinforces the possibility of food rejection at this level. Moreover, the few mucus cells at the anterior part could flush the surface of taste buds while their gradually increase caudally presumably slide the accepted food toward the stomach (Reifel and Travel, 1977).

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