CHAPTER 10

HISTOLOGICAL STUDIES OF THE ULTIMOBRANCHIAL BODY OF *HILSA ILISHA* AND *HILSA TOLI*

Since the beginning of the nineteenth century the ultimobranchial bodies, epithelial derivatives of the pharynx were studied in all the classes of vertebrates except the lower chordates and cyclostomes.(Rasquin and Rosenbloom, 1954, as reviewed by Van Dyke, 1959; Sehe, 1960, '65). In fishes, Giacomini (1908, cf. Van Dyke, 1959) found that the ultimobranchial body consisted of a granular substance mixed with degenerated nuclei. Watzka (1933, cf. Van Dyke, 1959) noted thyroid like colloid in both elasmobranches and teleosts. After a long interval, Sehe (1960) applied techniques of radioautography to settle the contradictory results obtained by several workers and came to the conclusion that ultimobranchial bodies of teleosts and amphibians neither metabolize nor store iodine and that this structure can in no way be considered the thyroid gland. He also demonstrated that after thyroidectomy these bodies did not show hypertrophy. On the other hand, Van Dykes(1959) several experiments suggested that in mammals the ultimobranchial body may be under the control of thyrotropin or thyroid gland.

So far no attempt has been made to study the changes taking place in the ultimobranchial bodies of migratory fishes. The present investigation was undertaken with a view to throw some light on the histological changes and the secretory activities

of the ultimobranchial bodies in relation to the changes in salinity, changes in neuroendocrine system and degenerative changes of alimentary canal and other visceral organs. The changes in ultimobranchial body of migratory (anadromous) H. ilisha and non-migratory H. toli during different phases of life cycle were studied.

MATERIAL AND METHODS

Live fishes from the net were taken and sacrificed immediately. The entire oesophageal region was taken out, cut opened and fixed in Bouin's fixative. Sections of 5 μ were cut and stained with haematoxylin-eosin and Heidenhain azan stains.

RESULTS

The ultimobranchial body of the fingerling of H. ilisha captured from river:

The ultimobranchial body of the fingerling was situated in the connective tissues, ventrolateral to the oesophagus and is surrounded by little fatty tissue. It has a conical form and the pointed end is supplied with many blood vessels (Fig. 1). The entire gland was covered by a thick capsule consisting of muscle fibres. The fibres were running parallel to each other and were solidly packed. The nuclei of this region were spindle shaped and were well stained with haematoxylin. These fibres in pointed end surround a major vessel. The main gland was divided into two regions. The first region occupies approximately 3/4th of the

entire gland. This region was made of fibres arranged in the form of a net work and empty spaces were seen. The muscle fibres or the septa which divided this gland were more eosinophilic. Nuclei of the cells of this region were very small and oval in shape (Fig. 2). Except for the nuclei the second region was similar to the other in histological structure. The nuclei of the cells were densely stained with haematoxylin and were bigger in size (Fig. 3). They were oval shaped. They were loaded with fine chromatin material. A few follicles which were partially filled with faint eosinophilic homogenous non-granular colloid were also observed in the vicinity of the capsular region (Fig. 4). Many capillaries were observed in this region.

The ultimobranchial body of immature H. ilisha from sea:

The entire gland had changed into an oval structure (Fig. 5) and was surrounded by loosely arranged muscle fibres. The outer wall of oesophagus also covered it. The capsule was not so thick as observed in the fingerling. The nuclei of the capsule were spindle shaped. The fibres of the capsule were loosely arranged. In some regions, the fibres had formed wavy mass having empty spaces. Also they were found broken at many places, leaving nuclei scattered and wandering. A mass of fat cells was also seen surrounding the ultimobranchial body. Blood vessels were observed on either side of the gland. The septa inside the gland divided the latter into three zones (Fig. 6). These zones were not observed clearly in the fingerling. The small syncitial walled follicles were scattered throughout the gland. Majority of the follicles were small. A few fillicles showed

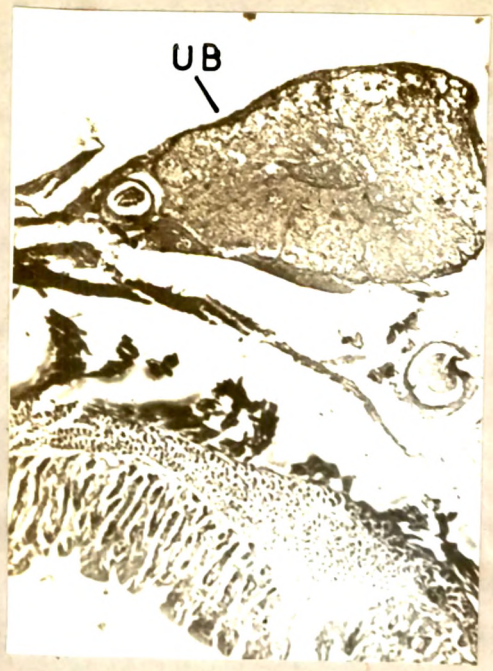

Fig. 1

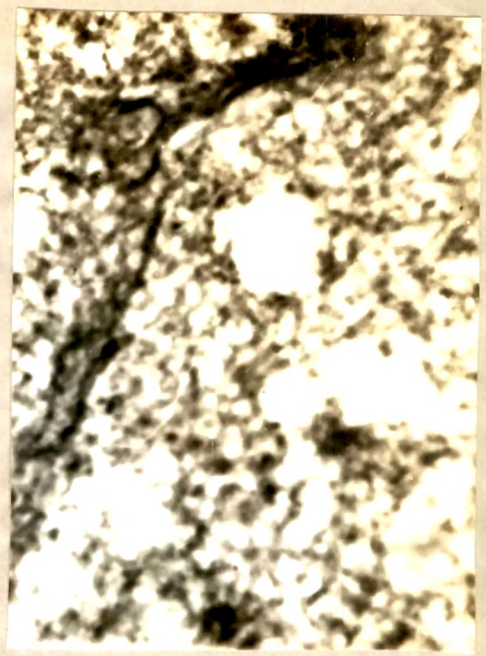

Fig. 2

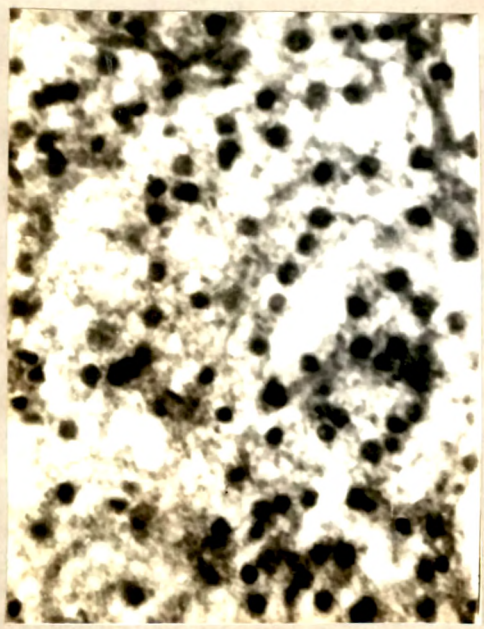

Fig. 3

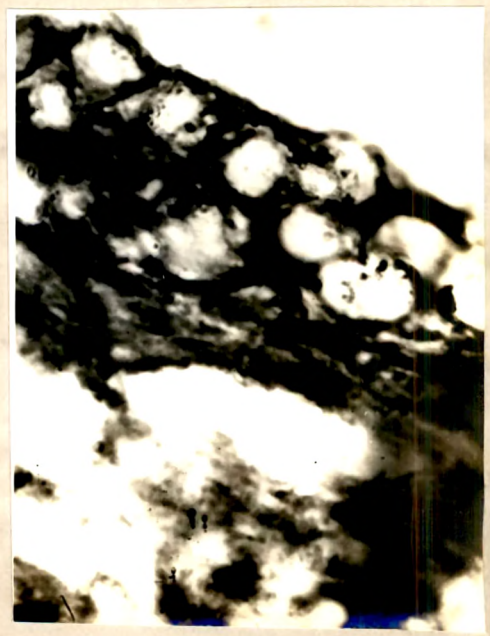

Fig. 4

Fig.1. Ultimobranchial body (UB) of fingerling of H. ilisha on the lateral side of the oesophagus. HE. X 63.
Fig.2. U.B. of fingerling showing fibres forming net work, small nuclei and empty spaces. Azan. X 1000.
Fig.3. U.B. of fingerling showing large, deeply stained nuclei. Azan. X 1000.
Fig.4. Follicles, near capsule in U.B. of fingerling, partially filled with colloid. Azan. X 1000.

an eosinophilic colloid which took brilliant red colour when stained with Heidenhain azan stain (Fig. 7). Very rarely a follicle or two showed aniline blue positive colloid giving blue colour.

The ultimobranchial body of mature H. ilisha during spawning, from river:

The perfect oval shape was not retained. A very thin layer of capsule persisted (Fig. 8). The muscle fibres of the capsule were arranged very loose and at many places they were observed to be broken leaving empty spaces. The nuclei of the muscle fibres retained their spindle shape and did not show pycnosis, but at the broken region the nuclei were found to be destroyed, shrunken and scattered in empty spaces.

The gland appeared to be active. Many follicles of small size alongwith bigger follicles were observed. These follicles were found throughout the gland and the different zones as observed in immature H. ilisha captured from sea were not so clearly visible. When stained with Heidenhain azan stain, the colloid in smaller and big follicles took up a brilliant red colour giving a homogenous appearance to it. In a few of the bigger follicles, the colloid took up bluish colour too. When stained with haematoxylin-eosin, the colloid in the smaller and a few big follicles showed more affinity for eosin whereas the remaining bigger follicles showed faint eosin staining. In a few follicles very little quantity of colloid was observed (Fig. 9). Here the colloid was found to be shrunk and a few granules were also seen. Some of the follicles were shrunk and were devoid of colloid. The septa dividing the gland was very thin and broken

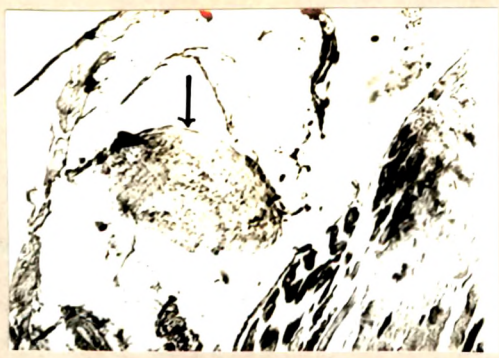

Fig. 5

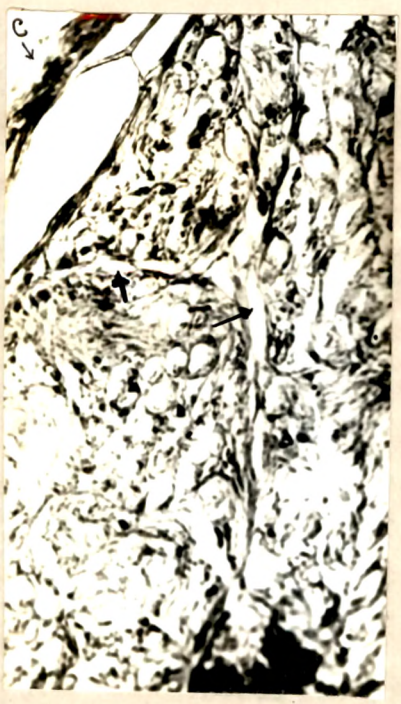

Fig. 6

Fig. 7

Fig.5. U.B. (arrow) of immature H. ilisha. Azan. X 63.

Fig.6. U.B. of immature H. ilisha showing septa (arrow) dividing the body into three regions showing follicles. C- capsule. Azan. X 400.

Fig.7. A few bigger follicles in U.B. of immature H. ilisha showing colloid. Azan. X 1000.

at many places probably due to overactivity of the follicles.

Summing up, the follicles had increased in number and size as compared to those of immature H. ilisha captured from sea and possessed a greater amount of homogenous colloid. It is evident that during maturity and migration the follicles of the ultimobranchial body showed characteristic changes in structure which indicate secretory activity. A few smaller (three to four) ultimobranchail bodies were also seen in the vicinity of the main gland the follicles of which were full of colloid. These bodies were similar in structrue to the main gland and showed similar secretory activity.

The ultimobranchial body of spent H. ilisha captured from river during remigration:

The shape of the ultimobranchial body was similar to that of spawning mature H. ilisha. The capsule had become comparatively thick (Fig. 10). The muscle fibres of the capsule were loosely arranged and were found to be shrunk at some places. They were wavy in appearance and due to shrinkage empty spaces were also seen. In such places nuclei were found scattered, separated from cytoplasm. Otherwise the nuclei retained the spindle shape and also showed chromatin material. However, in some regions the nuclei were enlarged and showed pycnosis. The septa which is a continuation of the surrounding capsule were found to penetrate deep inside the gland proper and divide the gland into 6 to 8 small areas (Fig. 10). The septa were similar in nature to the capsule and were very much thickened in spent H. ilisha in comparison with those of H. ilisha of various stages described above.

The nuclei of the muscle fibres of the septa were large in size and also showed pycnosis.

In the gland proper the follicles had shrunk and a few smaller follicles had appeared. Scattered among follicles, big nuclei stained brilliant red with Heidenhain azan stain were seen. The larger follicles, when stained with Heidenhain azan stain, showed the presence of a bluish colloid shifted to one side of the follicle, A few shrunk follicles, with a wavy follicular wall contained a slightly pinkish granular colloid, shifted to one side (Fig. 11). The smaller follicles also contained colloid. However, many follicles were found to be completely devoid of colloid or granular mass.

Two to four small ultimibranchial bodies were also observed in the near viscinity of the main gland (Fig. 10). These smaller bodies had a distinct thick capsule similar to the one surrounding the main ultimobranchial gland. These capsules showed a wavy appearance and loosening of fibres. Small follicles without colloid were found in the majority of these bodies. A few of the follicles showed shrunk and granular mass within (Fig. 12).

All the glands were found to be surrounded by the fatty mass at this stage also. On the whole it may be concluded that the glands were in an exhaust phase.

<u>The ultimobranchial body of non-migratory immature H. toli (stage II-III) from sea</u>:

The entire gland was oval shaped, covered by an outer

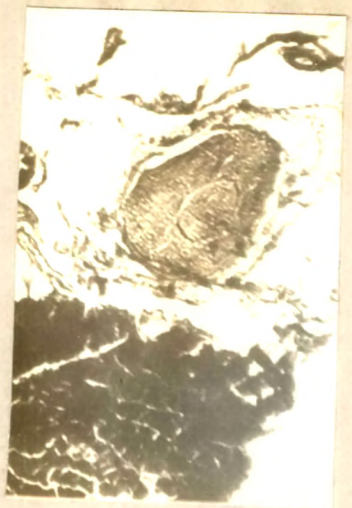

Fig. 8

Fig. 9

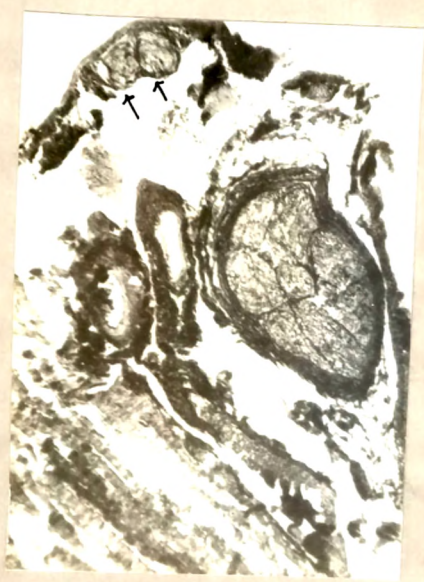

Fig. 10

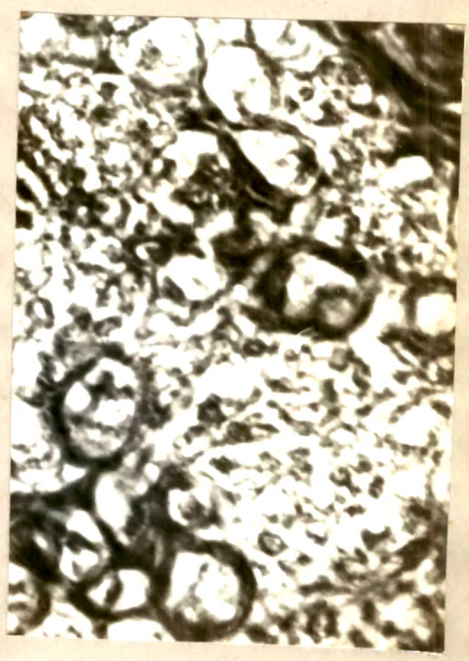

Fig. 11

Fig.8. U.B of mature H. ilisha. Note capsular degeneration. Azan. X 63.
Fig.9. U.B of mature H. ilisha showing a few follicles with little shrunk colloid. Azan. X 1000.
Fig.10. U.B of spent H. ilisha . Note (i) the thick capsule (ii) septa dividing the gland into small areas (iii) two new small bodies (arrow). Azan.X63.
Fig. 11.U.B of spent H. ilisha showing shrunk follicles with colloid shifted to one side. Azan. X 1000.

wall of the oesophagus. The capsule was moderately thick and the muscle fibres were more or less loosely arranged. At very few places the muscle fibres were seen broken and some regions only showed wavy nature of muscles. The nuclei of the muscle cells were spindle shaped. The septa penetrated deep inside the gland dividing the latter into three zones. Small follicles with syncitial walls were observed. In a few of the follicles a homogenous eosinophilic colloid which took up a red colour when stained with Heidenhain azan stain was observed. Rarely, in very large follicles aniline blue positive, blue coloured colloid was visible.

Fatty mass of cells was also found surrounding the ultimobranchial body. Many blood vessels were also seen around this region. In short, ultimobranchial body of the non-migratory immature H. toli was similar in all respects to that of migratory immature H. ilisha captured from sea.

The ultimobranchial body of mature H. toli captured from sea:

The ultimobranchial body was similar to that of mature migratory H. ilisha captured from river during spawning. The capsule was thin layered, consisting of loosely arranged muscle fibres. They were found broken at many places leaving the nuclei scattered. The nuclei were spindle shaped and filled with finely granular chromatin material. They were found shrunk where muscle fibres were destroyed. The division of the gland was not prominent as the septae were not thick.

The main gland exhibited an active phase. Many large

follicles were filled with homogenous colloid, staining brilliant red with Heidenhain azan stain. Small follicles were also observed with the colloid staining brilliant red with Heidenhain azan stain. A few follicles showed aniline blue positive, shrunk colloid whereas some showed pinkish colloid with granules. In haematoxylin-eosin staining the colloid showed varied eosinophilic nature. In smaller folliclesand in many of the larger follicles the colloid was strongly eosinophilic whereas in the rest it was less eosinophilic.

The ultimobranchial body of spent H. toli captured from sea:

The ultimobranchial body almost in all respects resembled that of spent H. ilisha captured from river. The entire gland was some what oval in shape, surrounded by a thick capsule consisting of muscle layers which were very loosely arranged and at some places broken also. Spaces were also seen in between the muscle fibres. At some places the muscle fibres were compactly arranged. The nuclei were spindle shaped and were densely stained with haematoxylin. Muscle fibres were found to be destroyed with their nuclei shrunk and scattered in empty spaces. Some times the nuclei were loosely attached to the remnants of destroyed muscle fibres.

The septa was found to penetrate deep inside the gland proper and divide the gland into 7-8 zones. The entire ultimobranchial body was conical in shape and the pointed region was supplied with blood vessels. The inner divided zones were shrunk and separated from the septa dividing the gland. The muscle

fibres were found broken at many places. The nuclei were large and scattered throughout the gland.

The follicles of the gland were filled with homogenous colloid shifted to one end. It stained blue with Heidenhan azan and faint pink with eosin. Some follicles showed clear, eosinophilic, homogenous colloid. However, many of the follicles were shrunk and devoid of colloid.

Three to four smaller ultimobranchial bodies were observed near the main gland. These ultimobrancial bodies resembled the main gland in structure and staining affinities.

The ultimobranchial body of drifted H. toli captured from river:

The capsule surrounding the main gland was thick walled, consisting of very loosely arranged muscle fibres, broken at many places (Fig.13) and leaving the nuclei scattered in empty spaces. The nuclei were spindle shaped. The inner septal region was represented by empty spaces in many places (Fig. 14), where ocasional remnants of destroyed muscle fibres were often observed.

The entire gland appeared shrunk, as if detached from the capsule. The staining affinity for haematoxylin-eosin as well as Heidenhain azan was much less. The majority of the follicles appeared to be devoid of colloid, loose and shrunk. In a few follicles faint pink granular colloid was seen (Fig. 15), when stained with Heidenhain azan stain. Surrounding the major gland there were three to four small glands with the capsule showing the same type of degenerative changes as that of the major gland. When stained with Heidenhain azan stain, small

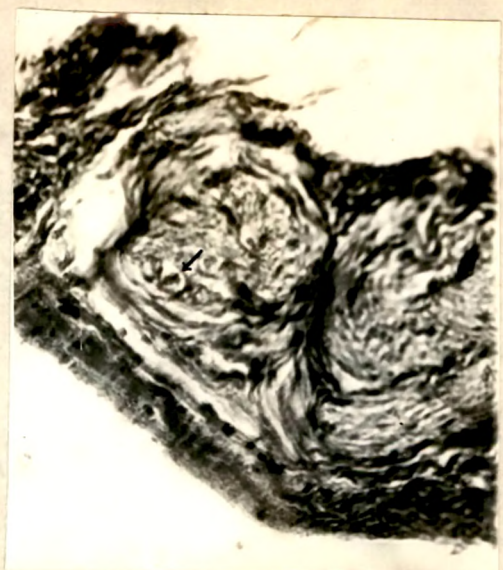

Fig. 12

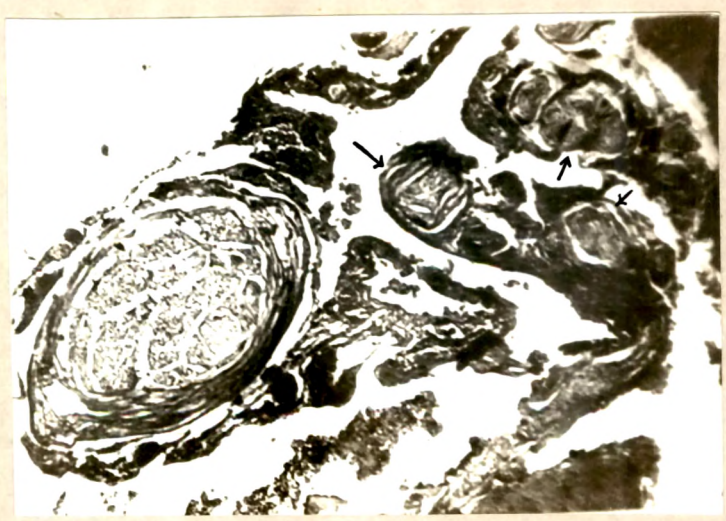

Fig. 13

Fig. 12. Small U.B of spent H. ilisha showing thick capsule and a few follicles with shrunk colloid (arrow) Azan. X 400.

Fig. 13. U.B of drifted H. toli. Note broken capsule and small additional bodies (arrow). Azan. X 63.

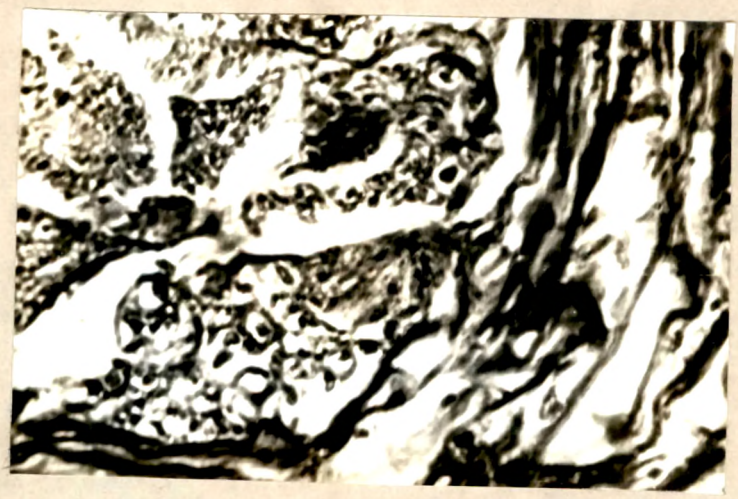

Fig. 14

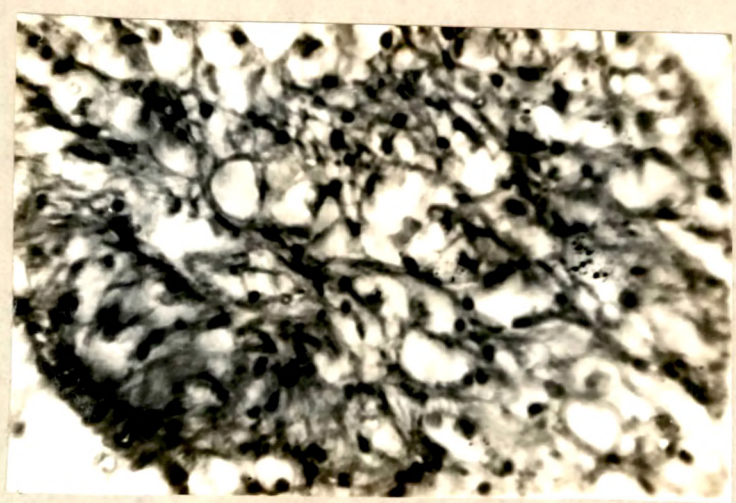

Fig. 15

Fig. 14. U.B of drifted H. toli showing degenerative changes. Note capsule wall and gland showing empty spaces. Azan. X 400.

Fig. 15. U.B of drifted H. toli showing follicles as described in the text. Note enlarged nuclei. Azan. X 630.

follicles showed uniform red colloid. The larger follicles were loose and contained granular red colloid, shifted to one end of the follicle. However, some of the larger follicles showed homogenous aniline blue positive colloid shifted to one end of the follicle.

DISCUSSION

In mammals and reptiles, the ultimobranchial body appears to be under control of thyroid gland. Van Dyke (1959) showed that thyrotropine as well as thyroid gland itself exerted an influence on ultimobranchial gland, and suggested that this gland may act as growth centre in mammals. Eggert (1938, cf. Van Dyke, 1959) noted an increase in the activity of ultimobranchial gland in reptiles when the animals were given TSH injections in the absence of thyroid tissue. However, in fishes it is known that thyroid gland never comes in contact with ultimobranchial gland as in mammals and it is established that ultimobranchial gland does not metabolise or store iodine and is not influenced by hypophysectomy or thyroidectomy (Sehe, 1960). It is obvious, therefore, that in fishes, the ultimobranchial gland may not be regulated by thyrotropin or thyroid gland.

It is interesting to note in the present study the secretory activity of ultimobranchial gland was found to be the maximum whenever the thyroid activity was at its peak eg. mature migratory H. ilisha and mature non-migratory H. toli captured from sea. Similarly when the activity of the thyroid gland

regressed, the ultimobranchial gland also showed a decrease in activity eg. spent H. ilisha and spent non-migratory H. toli. It may also be mentioned here that the non-migratory H. toli thyroid exhibited a goitorous condition (Desai, 1967) when drifted into iodine deficient river waters. The ultimobranchial body of the same fishes showed pronounced degenerative changes. The degenerative changes of ultimobranchial body may be considered to be due to the inability of the gland to cope up with the functional demands.

The above observations suggest a possible relationship between the activities of the thyroid and the ultimobranchial glands. However, since other factors like stress due to starvation, maturity and changes in salinity etc, also play an important role in bringing about morphological and neuroendocrine changes, it is difficult to say that only thyroid gland exerts its influence over the ultimobranchial body.

The present studies also show that along with age, the secretory activity of the ultimobranchial gland also changes. During maturity the secretory activity was observed to be at its peak eg. mature H. ilisha and H. toli. In the spent the secretory activity was regressed as revealed by the degenerative changes described. In the fingerling, there are very few follicles showing secretory activity. From these observations it may be concluded that the activity of the ultimobranchial gland is related to the growth of the animal. Eggert (1938, cf. Van Dyke, 1959) also showed that in reptiles the ultimobranchial body showed a progressive change from a cord like to a well vascularised and

thyroid like follicles during growth. These observations lend support to Van Dykes'(1959) observations that the ultimobranchial gland may act as a growth centre.