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STUDIES ON THE DEVELOPMENT OF THE OVIDUCT IN HIGH AND LOW EGG-PRODUCING FOWL
II -HISTOLOGICAL STUDIES

(With 1 table & 12 Figures)

BY

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(Received at 28/12/1992)

دراسات لتطور قناة البيض في الدجاج ذات الإنتاج العالى والمنخفض

٢ - دراسات هستولوجية

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استخدم في هذا البحث ستة وستون دجاجة من سلالتى الدندراوى والهاى لاين. تم تجميع العينات فى الفترة ما بين عمر يوم حتى عمر ٢٤ أسبوع بعد التفريخ.

ظهرت عملية تكوين الاهداب فى الخلايا المبطنة لقناة البيض من اليوم الاول بعد التفريخ فى كل من السلالتين وقد أمكن مشاهدة أربعة مراحل فى عملية تكوين الاهداب هى المرحلة القاعدية للسيتوبلازم، مرحلة ظهور الاجسام القاعدية، مرحلة ظهور الاهداب القصيرة، ومرحلة ظهور الاهداب الطويلة.

تميزت الخلايا العمادية المبطنة لقناة البيض الى خلايا مهدبة وخلايا غير مهدبة مفرزة فى عمر ٢٠ أسبوعا فى سلالة الدندراوي و٢٤ أسبوعا في سلالة الهاي لاين.

ظهرت بشائر غدد قناة البيض على هيئة أنابيب خلوية قصيرة فى قلب الثنايا المخاطية حيث كانت متصلة بخلايا النسيج الطلائى المبطن لهذه القناه، وقد أمكن مشاهدة هذه البشائر عندما بلغ عمر الطائر ٦ أسابيع فى مناطق عنق القمع والبرزخ والمهبل وعند ١٢ أسبوع لمناطق المعظم والرحم، وبتقدم العمر أخذت هذه الانابيب الغدية فى النمو والتفرع حيث أظهرت نشاطا افرازيا عند عمر ٢٠ أسبوعا فى سلالة الدندراوى و٢٤ أسبوعا فى سلالة الهاى لاين.

ظهرت الطبقة العضليه لقناة البيض بشكل واضح ابتداء من الاسبوع السادس على هيئة طبقة رفيعة من العضلات الملساء مرتبة ترتيبا دائريا وبتقدم العمر ازدادت هذه الطبقة فى السمك، وقد تميزت الى طبقتين طبقة داخلية مرتبة ترتيبا دائريا وأخرى خارجية مرتبة ترتيبا طوليا.

DEVELOPMENT OF THE OVIDUCT IN FOWL

SUMMARY

The present investigation was carried out to study the post - hatchery developmental changes in the oviduct of high (Hy-Line) and low (Dandrawi) egg-producing fowl within the period from the first day to 24 weeks after hatching. The epithelial lining of the oviduct of immature chickens showed clear evidence of ciliogenesis which occured sporadically at the first day after hatching. Four different morphological stages could be recognized within this lining epithelium. At the 20th week, the lamina epithelialis in Dandrawi was higher and more differentiated than that of Hy-Line breed. It was formed of ciliated and non ciliated (secretory) cells. These morphological features were demonstrated only at 24 weeks in Hy-Line breed. The newly formed oviducal glands at the infundibulum, isthmus and vagina were recognized earlier (6 weeks) than those at the magnum and uterus (12 weeks). They were represented by short tubules connecting the lamina epithelialis. With the advancement of age the lamina propria of the mucosal folds was occupied completely by the tubular glands at 20 weeks in Dandrawi and at 24 weeks in Hy-Line breeds. The muscular tunic was ill-developed at the earlier stages of development where it was represented by isolated smooth muscle fibers arranged in a circular manner. At later stages of development the muscular tunic was differentiated into an inner circular and an outer longitudinal smooth muscle layers.

INTRODUCTION

The avian oviduct consists of five distinct regions. Each region possesses its characteristic morphology and function *(RICHARDSON, 1935; RAMANOFF and RAMANOFF, 1949).*

A tramendous change in the size of the avian oviduct occurs during the reproductive cycle (developing, laying and regressing stages). These physiological processes are directly mediated through the action of gonadal hormones *(BRANT & NALBANDOV, 1956; PALMITER & WRENN, 1971 and YU & MARQUARDT, 1973 a).*

The cyto-differentiation of the oviducal mucosa, time of formation and the way of development of the oviducal glands have received little attention. Therefore, this investigation was carried out to illustrate the histological pattern of the oviduct in Dandrawi and Hy-Line as a low and high egg-producing breeds,

respectively; throughout the developmental stages from the first day to 24 weeks after hatching.

MATERIAL and METHODS

The present investigation was conducted at the Poultry Research Farm, Animal Production Department, Faculty of Agriculture, Department of Anatomy and Histology, Faculty of Veterinary Medicine, Assiut University, from Jan. to Dec. 1991.

The experimental breeds were used, one Egyptian local strain (Dandrawi) and one foreign breed (Hy-Line).

During the growing period (brooding and rearing periods) chicks were reared in batteries, provided with electrical heaters and air conditions to maintain the adequate temperature, up to sexual maturity.

At 20 weeks of age (prior to sexual maturity), pullets from each breed were distributed in individual cages under the same environmental conditions (25°C and 60-70% relative humidity). A growing diet containing about 18-19% protein and ME of 2817 Kcal/Kg was provided. The principal constituents of layers diet were 18% protein, 3.25% Ca, 0.8% P. Food and water were supplied ad libitum.

Regarding the lighting program used in this experiment, it is worthy to mention that one day old chicks were exposed to 24 hours light/day during the first week and then followed by 14 hours continuous photoperiod till the 21th week of age and 17 hours light/day from the 21th week of age till the end of the experiment.

Age and number of birds used in this experiment are shown in Table (1). Birds from each breed were slaughtered at 11:00 a.m.

The oviduct was immediately obtained after slaughtering and small pieces from each segment were fixed in Bouin's fluid. After proper fixation, the materials were dehydrated and embedded in paraffin wax. Step serial sections were cut at about 5-7 um.

The following stains procedures were adopted:

1- Harris's hematoxylin and eosin for general histological examination *(HARRIS, 1898)*.
2- Crossmon's Trichrome stain for demonstration of collagen fiber, smooth muscle, fuchsinophilic and orangophilic substances *(CROSSMAN, 1937)*.
3- Periodic Acid Schiff technique (PAS) for demonstration of neutral mucopolysaccharide *(McMANUS and MOWERY, 1960)*.
4- Alcian blue (ph= 2.5) for demonstration on acidic mucopoplysaccharide *(McMANUS and MOWERY, 1960)*.

Other small pieces from different segments of the oviduct were fixed in cold 5% glutaraldehyde. After dehydration, tissues were embedded in Epon, sectioned at 1 um thickness with LKB ultramicrotome and stained with toluidine blue and Alcian blue PAS technique.

Table 1: Material available in the present study

Age	No. of birds	
	D	H
1 day	3	3
1 week	3	3
2	3	3
3	3	3
4	3	3
6	3	3
8	3	3
12	3	3
16	3	3
20	3	3
24	3	3

D= Dandarawi H= Hy-Line

RESULTS

The wall of the oviduct consisted of three layers namely; an internal mucous tunic and an external serous or adventitial tunic (Fig. 1).

At the first day after hatchnig, the mucous tunic was thrown into few short longitudinal folds at the magnum, uterus and vagina (Fig. 2). The mucous tunic was represented by a single layer of epithelial cells resting on a highly cellular connective tissue lamina propria.

The lamina epithelials was of the simple columnar or the pseudostratified columnar variety (Fig. 2 & 3). The lamina epithelialis was invaginated at various intervals to form shallow epithelial grooves or crypts. The lamina epithelialis showed clear evidence of ciliogenesis. Ciliogenic cells were first recognized by the appearance of cytoplasmic basophilia at the supranuclear region of the columnar cells lining the oviduct. At the later stage of ciliogenesis, the ciliogenic cells demonstrated basal bodies at the free apical border of the columnar cells under the cell membrane. However, the free apical part was devoid of cilia. Other ciliogenic cells demonstrated very short and few cilia at the free apical border of the cell. The differentiated ciliated cells were recognized by the presence of abundant long ciliary shafts (Fig. 4). The subepithelial connective tissue lamina propria was highly cellular. The muscular tunic was ill-developed and represented by isolated smooth muscle fibers arranged in a ciruclar manner.

At 1-4 weeks, the mucous tunic of the tubular part of the infundibulum and the isthmus was thrown into small and short longitudinal folds. However, the mucosal folds of the magnum, uterus and vagina were increased in height and number than at the previous age.

At 6 and 8 weeks, the epithelial groovs became deeper and more branched. The ciliogenic cells of the lamina epithelialis wer more abundant than those observed at the previous groups (Fig. 5). The lamina epithelialis was invaginated at the tops, sides and base of the mucosal folds at the infundibulum; isthmus, and vagina to form the elements of the oviducal glands. The newly formed glands were represented by short tubules connecting to the lamina epithelialis (Fig. 6). The lamina propria was formed of dense connective tissue containing smooth muscle fibers and small blood vessels. The muscular tunic became thicker and formed of circularly arranged smooth muscle fibers.

At 12 and 16 week, the ciliogenic cells were more numerous in Dandrawi than in Hy- Line breed. The epithelial grooves became deeper and more branched than those described at the previous stage. The epithelium lining the magnum and uterus was invaginated at tops, sides and base of the mucosal folds to from the magnal and uterine tubular glands, respectively. The newly formed glands were represented by short tubules connecting to the lamina epithelialis (Fig. 7). The oviducal glands at the infundibulum isthmus and anterior vagina were better developed than those observed at the aforementioned group. They were lined by 6-8 Low columnar cells with rounded basaly situated nuclei. The cytoplasm showed no secretory activity. At the isthmal segment the oviducal glands were better developed in Dandrawi than in Hy-Line breed. At the uterine segment the muscular tunic could be differentiated into an inner circular and outer longitudinal layers.

At 20 weeks, the mucosal folds in Dandrawi were markedly increased in number and height than those in Hy-Line breed. At the isthmal and vaginal segments, the mucosal folds were long and cylinderical in shape with additional numerous small secondary folds (Fig. 8). The lamina epithelialis in Dandrawi breed differentiated into two types namely; ciliated and non ciliated (secretory) cells (Fig. 9). The ciliated cells were wider and tapered towards the basement membrane. Their nuclei were Large, spherical, vesicular and Located at the apical part of the cells. The cytoplasm contained considerable amount of supranuclear basophilic granules which varied in size and number from cell to cell. The cytoplasmic granules were negatively stained with Alcian blue or PAS technique. The non ciliated (secretory) cells were more cylindrical with narrow apical part. Their nuclei were oval, vesicular and located towards the basement membrane. The cytoplasm contained small basophilic granules within the supranuclear region. These granules were PAS - Alcian blue positive (Fig. 10). Large vacuoles were found in both the luminal and basal sides of

the nuclei of these cells. The oviducal glands were relatively more developed in Dandarwi than in Hy-Line breed. These glands were of branched tubular type and their epithelium was of the pyramidal variety. The cytoplasm of the uterine glandular cells was lightly stained, however that of the magnal glands occupied large metachromatic secretory granules which reacted moderatly to Alcian blue and PAS-technique (Fig. 11 a & b). The infundibular gland in both Dandrawi and Hy-Line chickens occured only at the terminal part of the infundibulum near its junction with the magnum. The secretory granules of the infundibular glands were smaller, less numerous and denser than those of the magnal glands. They contained similar PAS reactive granules. At the vagina the oviducal glands were found only in the vicinity of the utero-vaginal junction. They were less coiled and less branched than those observed at the other segments of the oviduct. They were negatively stained with Alcian blue and PAS technique.

The muscular tunic was more developed in Dandrawi than in Hy-Line breed. It was differentiated into an inner circular and an outer longitudinal smooth muscle layers.

At 24 weeks, the changes observed in the oviduct of both breeds was more quantitative than qualitative. The lamina epithelialis in Hy-Line breed was differntiated into two types namely, ciliated and non ciliated secretory cells bearing the same characteristics described for the two types in Dandrawi at the 20th week.

The oviducal glands at both the magnal, isthmal and anterior vaginal segments of Hy-Line breed increased markedly in amount as more as in Dandrawi breed (Fig. 12 a & b).

DISCUSSION

The present study revealed that the oviducal epithelium in both Dandrawi and Hy-Line breeds, being of the simple columnar or pseudostratified columnar variety, showed clear evidence of ciliogenesis which occured sporadically in immature oviduct and was prevalent in fully differentiated oviduct at 20 weeks in Dandrawi and 24 weeks in Hy-Line breed.

The oviduct of the chick shares with that of several mammals the property that estrogen can stimulate ciliogenesis. *PALMITER* and *WRENN (1971)* and *SEGAL (1974)* noted that estrogen stimulated ciliogenesis in the magnum portion of immature chick oviduct. *ANDERSON and HEIN (1976)*, studied the morphological details of estrogen dependent ciliogenesis in the shell gland of the chick oviduct and mentioned that the rate of ciliogenesis was found to be affected by progesterone and type of estrogen administered. With respect to the function of the ciliary shafts of ciliated cells *ROMANOFF and ROMANOFF (1949)* and *DRAPER; JOHSTON and WYBURN*

(1968) postulated that they were required to facilitate the release of the secretory material from the secretory cells.

The present study revealed that the oviducal epithelium was differentiated into ciliated and nonciliated (secretory) cells> The possibility of transformation of one type of cells to another type was recorded by some authors. RICHARDSON (1935) stated that the proportion of non-secretory ciliated to secretory non-ciliated cells on an epithelial surface may vary from time to time as the result of the transformation of one to another type and this occurred regularly in the oviduct of the fowl, the proportion of the secretory cells increasing as the time of ovulation approached. ARNOLD and SHOREY (1985), reported that the cells lining the oviduct of the possum are multipotential and that ciliated cells may arise as a result of redifferentiation of existing secretory cells. The secretory cells of the lamina epithelialis in both Dandrawi and Hy-Line showed PAS-positive material and were negatively stained with Alcian blue. They failed to give metachromacia with Toluidine blue. According to ROBINSON; KING and BOWEN (1968) the isthmus appeared to be rich in neutral mucin which might be glycoprotein. HOFFER (1971) found that the granules of the secretory cells in the lamina epithelialis of the oviduct in Quail were composed of non-sulphated carbohydrate.

The present stduy shwoed that, at 20 weeks, the ciliated cells of the lamina epithelialis in Dandrawi breed contained supranuclear basophilic granules which varied in size and number from one cell to another. These granules were negatively stained with Alcian blue and PAS-technique. Moreover, the non-ciliated cells contained smaller basophilic granules which stained with PAS and Alcian blue. These morphological features were observed for Hy-Line breed at 24 weeks. The secretion of the proteinaceous shell matrix and cuticle would appear to occur in the surface epithelium. Shell matrix probably being secreted by both ciliated and non-ciliated secretory cells (BREEN and DeBRUYN, 1969) and the cuticle by the ciliated cells (JOHNSTON et al. 1963 and AITKEN, 1971). TURCHINI (1924) considered that the calcified shell is produced by the surface epithelium of the uterine pouch but RICHARDSON (1935) related shell mineral secretion to the tubular glands. this latter function has also been suggested by studies of JOHNSTON et al. (1963) and BREEN & DeBRUYN (1969). However, recent physiological studies have shown that the surface epithelium may be associated with calcium transport in the shell gland (GOY; CAROL and SCHRAER, 1971).

According ROMANOFF and ROMANOFF (1949) the Vagina is responsible for the formation of the outer most layer of the egg shell or cuticle layer which is mucous in character. RICHARDSON (1935) reported that the secretory cells of the lamina epithelialis of the uterine pouch was responsible for production of cuticle on the surface of the egg shell.

The present study revealed that the oviducal epithelium in both breeds was invaginated at tops, sides and bases of the mucosal folds to form elements of the oviducal glands. The newly formed glands were represented by short tubules locating under the surface epithelum. The infundibular, isthmal and vaginal glands developed earlier (6 weeks) than the magnal and uterine gland (12 weeks). *PROCHAZKOVA (1971 a)* who mentioned that the subepithelial newly formed glands appeared at 22 weeks of age in all regions of the oviduct in domestic fowl at the same time. *TECHVER (1965)* reported that the newly formed oviducal tubular glands were formed gradually in the caudocranial direction. Variation in the time of differentiation and appearance of newly formed gland might be attributed to the difference in the stimulatory effect of the ovarian hormones on the various segments of the oviduct. *OKA and SCHIMKE (1969 a,b)* found that administration of estrogen to immature female chick leads to rapid cell proliferation, formation of oviducal tubular glands. They added that withdrawal of the hormone resulted in a regression of the oviduct with involution of tubular glands.

It is clear from the present result that the infundibular glands in both Dandrawi and Hy-Line chickens occured only in the terminal part of the infundibulum near its junction with the magnum. The secretory granules of the infundibular glands were smaller, less numerous and denser than those in the magnum. They contained similar PAS reactive granules. The infundibulum and magnum can be regarded as a functional of most of the protein and some minerals of the white or albumen of the egg *(WARREN and SCOTT, 1935)*. The first albumen layer is deposited on the ovum in the infundibulum *(SCOTT and HUANG, 1941)*. As the egg descends the magnum, more albumen is deposited in a series of concentric layers *(SCOTT and BURMESLER, 1939)*.

Vaginal glands of both Dandrawi and Hy-Line breeds occured only in the anterior vagina in the vicinity of the utero-vaginal junction. They were less coiled and less branched as those observed in the other segments of the oviduct. They were negatively stained with Alcian blue and PAS-technique. These results were similar to that obtained by *(FUJII;* 1963; GILBERT; *REYNOLDS; LORENZ, 1968 b and MERO & OGASAWARA, 1970). PROCHAZKOVA (1971 b)* in white Leghorn hens, *EL-BARGEESY (1990)* in Turkey, *EL-HABBAK (1990)* failed to demonstrate the vaginal glands. *MERO and OGASAWARA (1970)* reported 2 categories of utero - vaginal sperm storage tubules in the oviduct of white Leghorn hens, tubules with large luminal area (enlarged tubule) and tubules with small luminal area (non-enlarged) tubular enlargement was associated with sperm release.

It is worthwhile to notice that the oviduct in Dandrawi breed reached an advanced developmental stage more earlier at 21

weeks than that of the Hy-Line breed. However, at the 24th week of age the developmental picture as well as the activities of the oviduct of the Hy-Line exceeded that of Dandrawi breed. This shwoed that Hy-Line breed required longer time to be matured. This may be attributed to the wide genetic variation in the age of their sexual maturity. *PEELET; GLAZENER and BLOW (1955)* mentioned that the onset of sexual maturity is the summation of morphological and physiological changes which culminate in the normal reproductive ability of the organism. The factors affecting the onest of sexual maturity may be either external factors as temperature, nutrition, day length, wavelength and intensity of light; or inherent physiological mechanisms. Not all hen come into lay at the same time when kept under apparently identical conditions suggesting there is a genetic factor involved. This characteristic has been exploited in attempts to develop strains maturing at an increasingly earlier age. Age at first egg is moderately heritable.

In conclusion, the oviduct in Dandrawi breed reached an advanced developmental stage more earlier (20 weeks) than that of the Hy-Line breed (24 weeks). However, at the 24th weeks of age of developmental picture as well as the activities of the oviduct of the Hy-Line exceeded that of the Dandrawi breed.

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LEGENDS

Fig.. 1: The magnum of Hy-Line breed at the 1st day after hatching, showing:
A- Mucous tunic. B- Muscular tunic.
C- Serous tunic.
(Haematoxylin and Eosin stain, oc. = x 6.3, ob.= X 25).

Fig. 2: The magnum of Hy-Line breed at the 1st day after hatching, showing the longitudinal mucosal folds.
A- Lamina epithelialis.
B- Connective tissue lamina propria.
(Haematoxylin and Eosin stain, oc. = X 6.3, ob. = X100).

Fig. 3: The isthmus of Hy-Line breed at the 1st day after hatching.
A- Lamina epithelialis.
B- Connective tissue lamina propria.
(Haematoxylin and Eosin stain, oc. = X 12.5, ob. = X 40).

Fig. 4: The infundibular epithelium of Hy-Line at the 1st day after hatching, showing the phenomena of ciliogenesis:
1- Cytoplasmic basophilia. 2- Basal bodies.
3- Short cilia. 4- Differentiated ciliated cells.
(Semithin Section, Toluidine blue stain oc.= X 10, ob.= X100).

Fig. 5: The lining epithelium of the magnum of Hy-Line breed at the 8th week after hatching, showing ciliated cells (C).
(Semithin section, Toluidine, blue stain, oc. = X10, ob.= X 100).

Fig. 6: The wall of the tubular part of the infundibulum of Dandrawi breed at the 6th week after hatching, showing the newly formed infundibular gland (G).
(Haematoxylin and Eosin stain, oc. = X = 6.3, ob.= X 40).

Fig. 7: The magnum of Hy-Line breed at the 16th week after hatching, showing the magnal glands (G).
(Haematoxylin and Eosin stain, oc. = X 8, ob. = X 100).

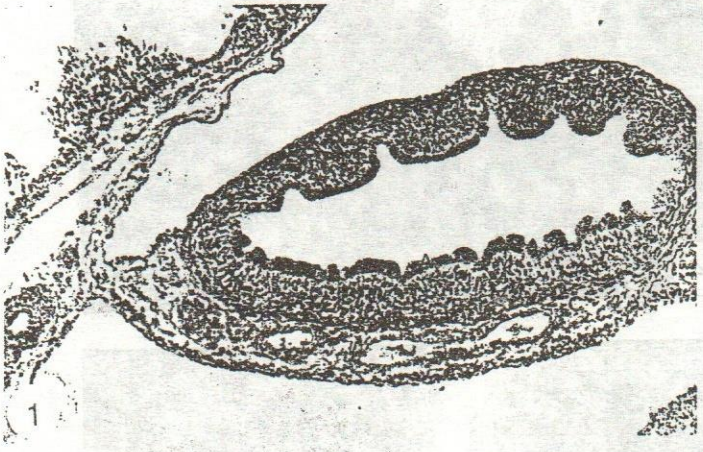
Fig. 8: The wall of the magnum at the 20th week after hatching.
a) Dandrawi breed. b) Hy-Line breed.
(a: Crossmon's Trichrome stain)
(b: Haematoxylin and Eosin stain).
(oc. = 6.3, ob. = 6.3).

Fig. 9: The lamina epithelialis lining the uterus of Dandrawi breed at the 20th week after hatching, showing the ciliated cells (C) and the non ciliated (Secretory) cells (S). (Semithin section, Toluidine blue stain, oc. = X 6.3 , ob. = X 100).

Fig. 10: The wall of the uterus of Dandrawi breed at the 20th week after hatching, showing PAS positive-granules within the epithelial secretory cells.
(PAS and Haematoxylin technique, oc. = X 8 ob. = X 100).

Fig. 11: The wall of the magnum of Dandrawi breed at the 20th week after hatching showing,
a- Various metachromatic secretory granules within the magnal glands (G).
b- Various PAS positive-secretory granules within the semithin sections, magnal glands (G).
a- Toluidine blue stain.
b- Alcian blue PAS technique, (oc.=X 10 ob.=X 100)

Fig. 12: The wall of the isthmus at the 24th week after hatching, showing the isthmal glands.
a- Hy-Line breed.
(Crossmon's Trichrome stain).
b- Dandrawi breed.
(Haematoxylin and Eosin stain)
(oc.= X 8, ob.= X 6.3).



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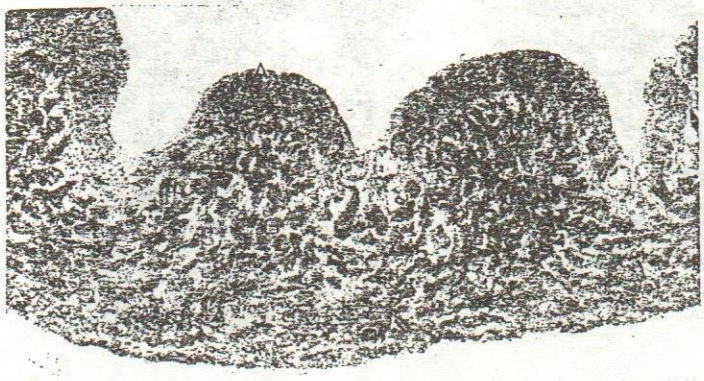

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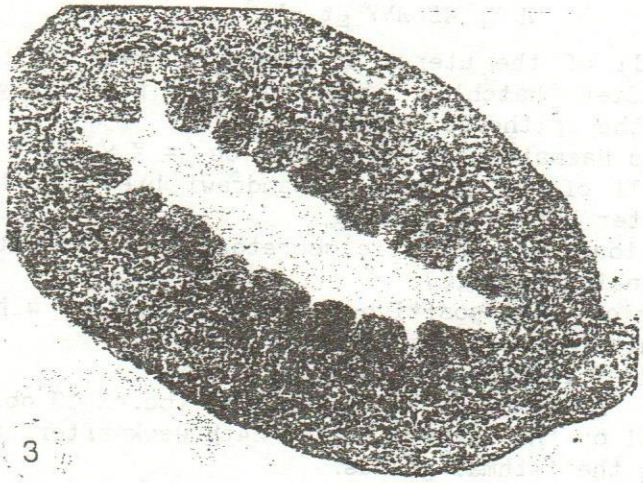

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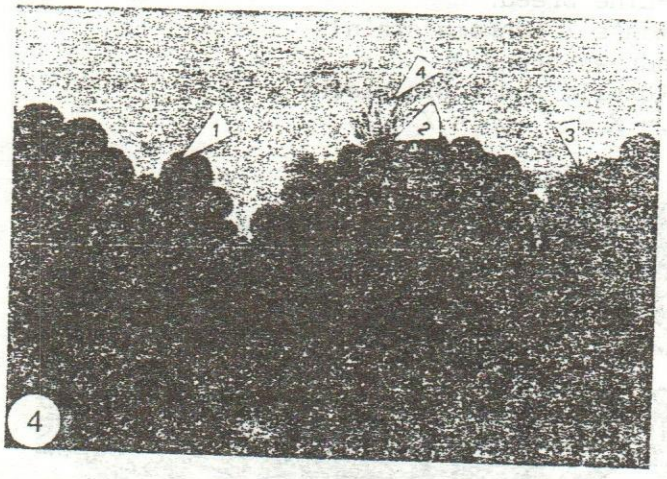

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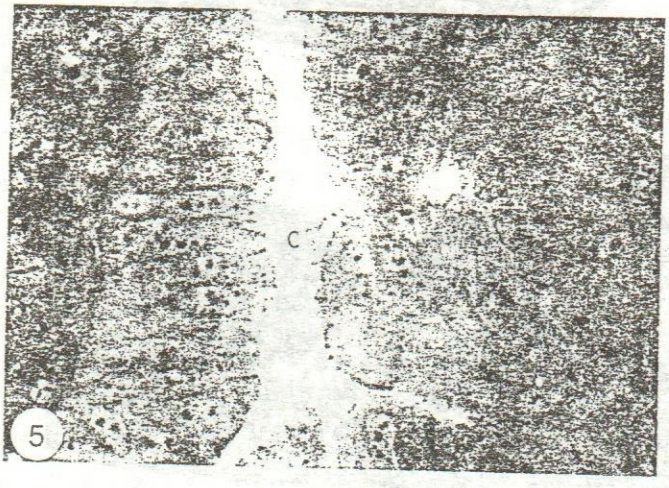

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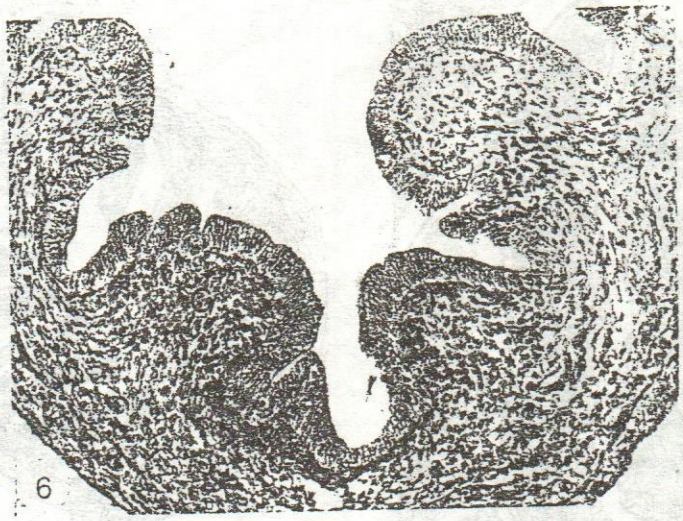

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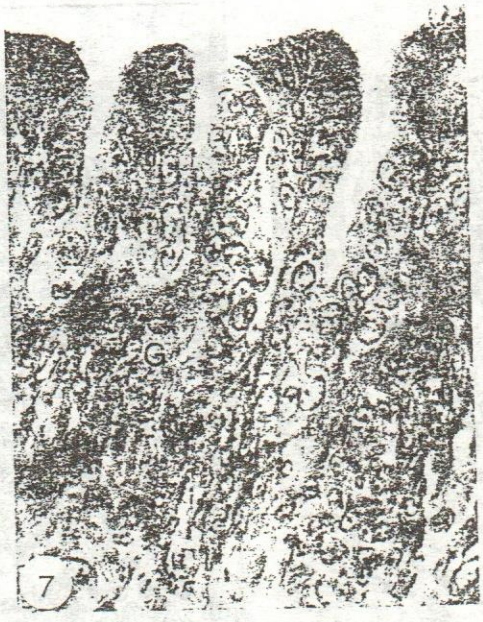

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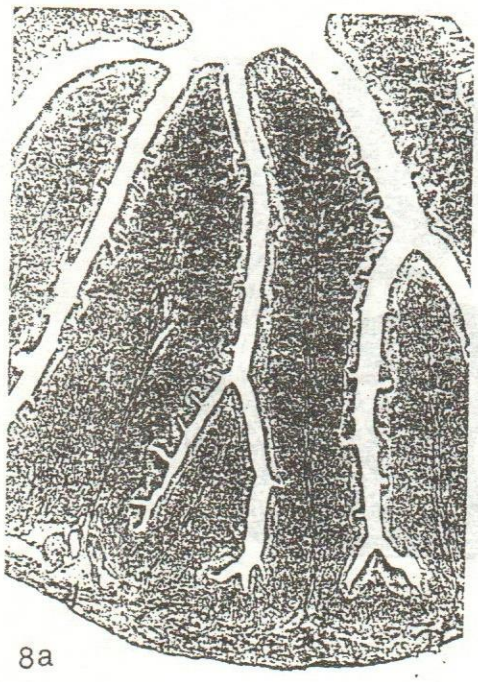

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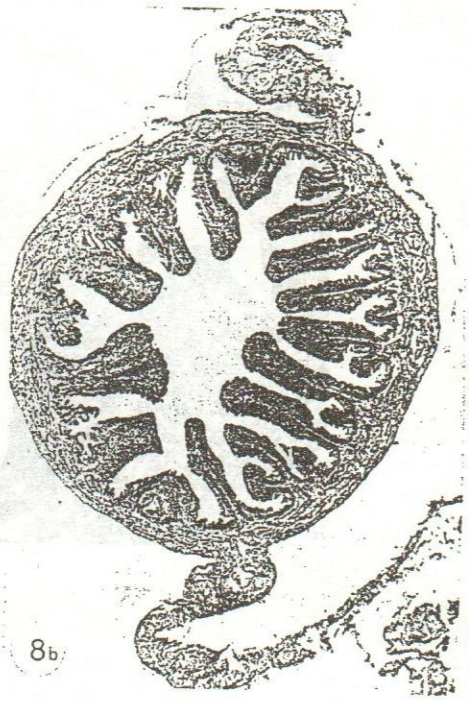

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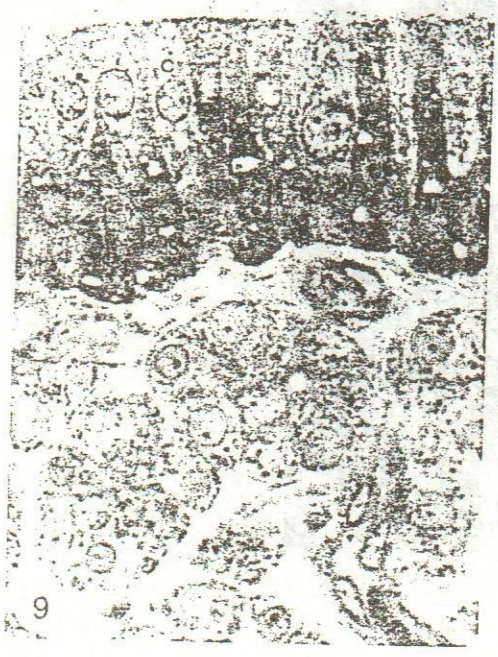

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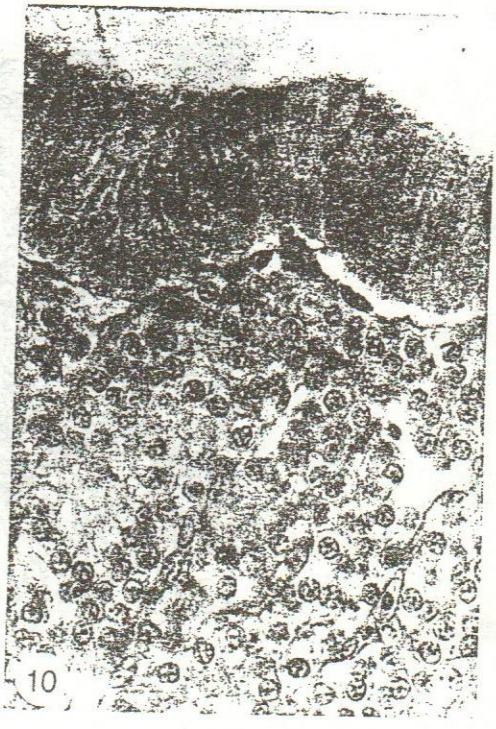

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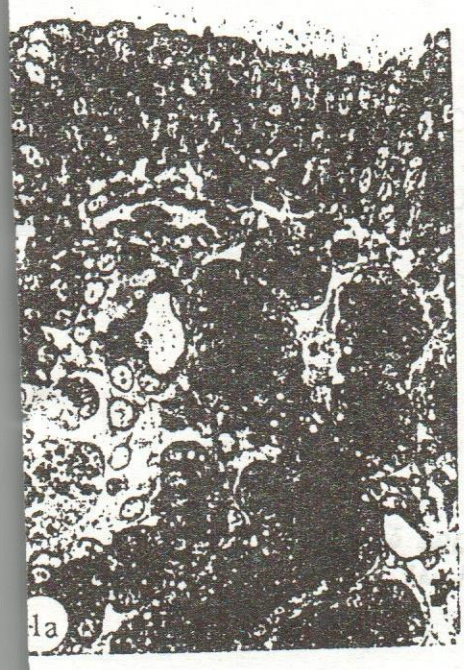

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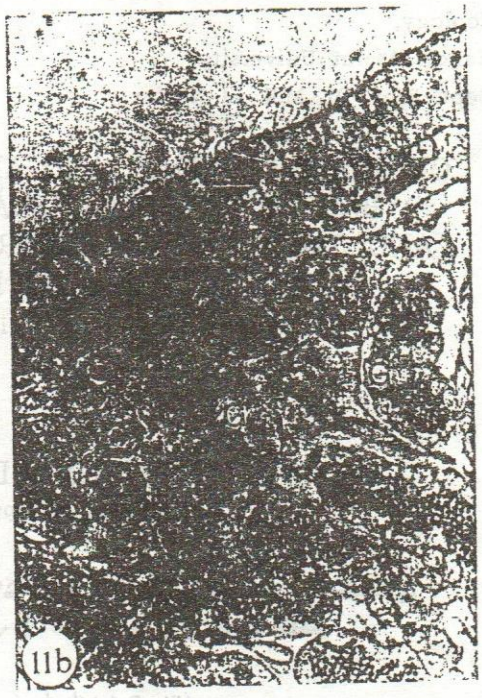

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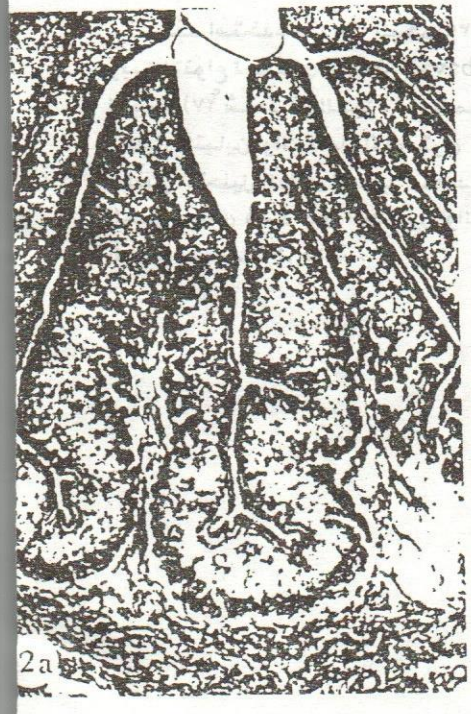

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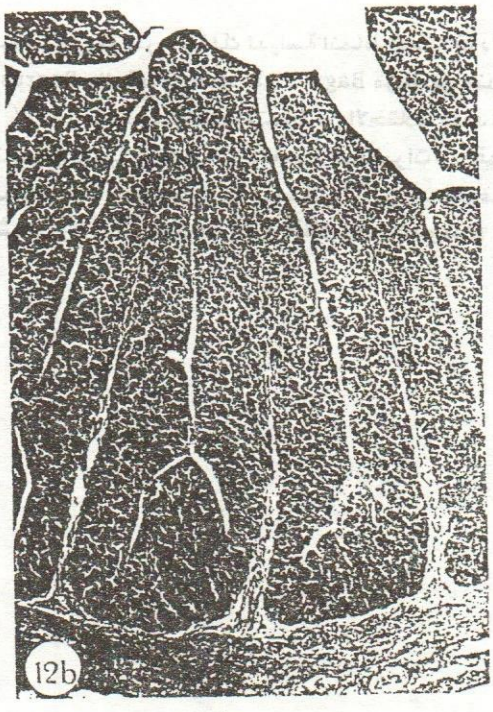

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