HISTOGENESIS OF MANDIBULAR SALIVARY GLAND IN NEWZEALAND WHITE RABBITS

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SUMMARY

The mandibular salivary gland was firstly appeared at 13 days old rabbit fetus as an epithelial buds invaginated from the linguogingival groove. At 15 days central cavities appeared in the developing buds converting them into tubules which later on branched and formed the developing adenomeres. As the development progressed the developing adenomeres increased in number and size and showing positive reaction for aldhide fuchsin and alcian blue stains. After birth the developing adenomeres became well advanced formed mainly of mucous and some mixed adenomeres. The gland reach its maximum development at 4 months of age where the adenomeres became fully developed giving strong alcianophilic reaction and the secretory cells showed large amount of electron lucent secretory granules and well developed Golgi complex. The duct system of the gland became well established

INTRODUCTION

The mandibular salivary gland is one of the major salivary glands. It produces substantial amount of saliva into the mouth.

The prenatal development of mandibular salivary gland has been studied by many authors in different animal species as rat *(Kim et al, 1970 and Yamashina and Barka, 1973)*, mouse *(Yohra, 1970; Borghese et al, 1974 and Ikehata, 1975)*, rabbits *(Emi, 1939 and Goto, 1959)*, pig *(Moral, 1913)*, buffalo *(Yadm, 1985)*, camel *(Abuzaid et al, 1990)* and domestic animals *(Elhagari, 1967; Noden and Lahunta, 1985 and Latshow,1987)*.

The mandibular salivary gland begins its embrylogical development just before or with the sublingual gland and becomes functioning before birth *(Chaudhry et al, 1983; Nagato et al, 1998 and Hakim et al, 2002)*.

The salivary secretion of mandibular gland is mainly mixed as the gland contains both serous and mucous secretory *units (Sivakumar et al, 2003 and Parthasarathy and Gopinathan, 2005)*.

The mandibular salivary gland as being one of the interesting organs requires a detailed description of its structural sequence of events. Lack of informations in this regard in rabbits was the stimulus for this work to be a basis for further studies.

MATERIALS AND METHODS

A total number of 20 Newzealand white rabbit fetuses ranged from 11-30 days of age were subjected to this study. In addition , 15 mandibular gland specimens of apparently healthy male and female rabbits aged 1-120 days were used . The head regions of fetuses and the gland specimens were fixed in 10% neutral formalin, Suza and Helley's fluid , processed and embedded in paraffin blocks.Cross and/or sagital stepserial sections of 4-6 um thick were cut and stained with Harris hematoxylin and eosin, Crossmon's trichrome stain, Aldehyde fuchsin stain and alcian blue stain as described by *Drury and Wallington (1980).*

For EM study , 1mm pieces of mandibular salivary gland at 29 days old fetus besides 2 weeks, 4 weeks and 4 months old rabbits were immersed in 3% glutraldhyde in 1M phosphate buffer (pH=7.3) for 2 hours, post fixed in cold 1M phosphate buffered 1% osmium tetroxide (pH=7.3) for 3 hours,and then briefly rinsed in distilled water *(Hayat,1986).*

Ultra thin sections were prepared , mounted on copper grids, stained in 5% uranyl acetate dissolved in 70% ethanol followed by lead citrate stain *(Reynold,1963)* and examined in a Joel 100CX transmission electron microscope at 60 KV.

RESULTS

The primordia of the mandibular salivary gland was firstly appeared at 13 days old rabbit fetus as solid epithelial buds invaginated from the linguogingival groove (Fig.1).Each bud was formed of closely packed cells having oval to rounded nuclei and lightly acidophilic cytoplasm. The budded cells had ill-distinct cell boundaries and merged directly without basal lamina with the surrounding undifferentiated mesenchymal tissue.

At 15^{th} day, the initial gland buds increased in number and extended deeply in the mesenchymal tissue. Central cavities began to appear in the gland buds converting them into elongated tubules with proliferating lining epithelium (Fig.2).

Upto the 17^{th} day the canalized tubules became branched and numerous separated by mesenchymal tissue (Fig.3). The developing gland was permeated by many blood vessels housing blood cellular elements in the mesenchymal tissue which represents the future stroma of the gland (Fig.4).

As the development progressed at the 20^{th} day of fetal life the duct system began to appear as long and branched intercalated duct lined by lightly stained cuboidal cells with central spherical nuclei (Fig.5). Also numerous cells were clustered at one side of the developing adenomeres representing presumptive site of prospective serous cap or demilune. On the other hand the rest of the cells were designated to form future mucous adenomeres. The mesenchymal tissue was still loose.

On reaching the 22nd day of fetal life, the developing adenomeres continued to increase in number on the expense of the mesenchymal interstitial tissue. Moreover, the amount, size and complexity of branching of ducts were enhanced (Fig.6). The connective tissue stroma arranged to form connective tissue trabeculae between the developing lobules.

At 24 days old fetuses, luminization of developing adenomeres became more advanced and large interlobular duct was more pronounced (Fig.7). The vacuolated cells were markedly increased in number with their characteristic flattened nuclei. Some acini showed positive aldhyde fuchsin reaction (Fig.8) while most of them showed moderate alcianophilic reaction (Fig.9). Ductal cells and lumina showed negative reaction for both stains.

From 26th day upto the full term fetuses, the mesenchymal interstitial tissue was greatly reduced and the capsule became well developed containing blood vessels and extended as septa between the gland lobules(Fig.10). The adenomeres became more developed and the proliferation of mucous cells with their flattened nuclei as well as their arrangement in a single layer against the basment membrane were enhanced. Some serous demilunes or seromucoid adenomeres became more pronounced. The mucous adenomeres still showed moderate alcianophilic reaction while the ducts were negatively reacted (Fig.11). The mucous secreting cells were studded with short microvilli on their luminal surface. The cytoplasm of these cells showed electronlucent mucous granules of different shapes and sizes (Fig.12). Extensive network of granular endoplasmic reticulum with dilated cisternae and rounded mitochondria with tubular cristae were also pronounced (Fig.13). desmosomal junction was found between the elongated myoepithelial cells surrounding the secretory adenomeres.

At 2 weeks old rabbits the secretory acini became more vacuolated and the mixed or seromucoid adenomeres became well established (Fig.14). The granular endoplasmic reticulum of mucous adenomeres became highly extensive with numerous dilated cisternae containing electronlucent mucoid materials (Fig.15). The cells of the serous demilune showed well developed rough endoplasmic reticulum and numerous rounded electron dense granules (Fig.16). At this stage strong alcianophilic reaction was noticed in both mucous adenomeres and luminaal contents of intercalated ducts (Fig.17).

As development advanced, few fat cells appeared between the mandibular acini of one month old rabbit. The acini became well developed , increased in size and formed mainly of mucous type beside some serous demilunes (Fig.18). the cells of the mucous adenomeres were high cuboidal to pyramidal in shape and had flattened to oval nuclei that situated against the base of the cell. Their cytoplasm appeared vacuolated, pale and foamy. The mucous granules became larger in size and increased in number while the luminal surface of the mucous cells still carrying short microvilli (Fig.19). The cells of the serous demilune showed spherical to oval nuclei and contained rounded

electron dense smaller granules (Fig.20). Also well appparent myoepithelial cells were noticed around the serous demilune.

On reaching 4 months old rabbit the mandibular gland reach its full maturation and maximum development. It was found that both inter and intralobular ducts were greatly increased in number and size together with the increase in the acidophilia of their lining cells. Moreover basal striation became well pronounced in the striated ducts which were lined by columnar cells with oval nuclei (Fig.21).The large interlobar duct was larger in diameter with wider lumen and slightly folded mucosa lined by tall columnar epithelium (Fig.22). The cytoplasm of mucous adenomeres was completely filled with large-sized numerous secretory granules while the cytoplasmic organelles constituted only a narrow zone around the flattened nucleus (Fig.23). Extensive alcianophilic reaction was noticed in the mucous adenomeres and luminal content of excretory ducts (Fig.24).

DISCUSSION

The present investigation revealed that the mandibular salivary gland primordia is originated as a bud from the linguogingival groove. Similar result was mentioned by William and Warwick (1980) and Sivakumar et al (2003) in man,, Latshow (1987) in domestic animals and Abuzaid et al (1990) in camel.

The postulations of Latshow (1987) that the close relationship of epithelial groove giving rise to the mandibular duct and gland with tongue which is essentially a pharyngeal structure leads to the suggestion that the gland is considered to be endodermal in origin.

At 15th day the initial gland bud extended deeper in the mesenchymal tissue. Central cavities began to appear forming elongated tubules which later on became numerous and branched forming the future secretory adenomeres. These results go hand to hand with those of Yohra (1970), Ikehata (1975), Abuzaid et al (1990) and Sivakumar et al (2003). Yadm (1985) in buffalo found that the secretory end pieces were lined by stratified epithelium while Nagato (1998) mentioned that the secretory adenomeres were lined by single layer of epithelial cells showing some stratification.

In agreement with the findings of Yadm (1985) the present study revealed that luminization of parenchymatous sigments was caused by separation and degeneration of their centrally located cells. On the contrary, Borghese (1950) in mouse attributed such process to the apart movement not to degeneration.

As the development progressed at 20 days cytoplasmic vacuoles began to appear and the vacuolated cells designated to form the future mucous adenomeres. Meanwhile, the presumptive site of the developing serous demilunes was recognized where some acidophilic cells started to be clustered in a cap-like around one side of the recently formed mucous adenomere. These findings agree with those of Borghese (1950), Yadm (1985), Abuzaid et al (1990), Nagato et al (1998), El-Bargeesy (2001) and Parthasarathy and Gopinathan (2005)

Our work revealed that the mandibular gland of the fullterm rabbit fetus became well developed, increased in size, vacuolation of its cells and diameter of secretory adenomeres. Similar results were mentioned by Yadm (1985), Abuzaid et al (1990) and Sivakumar et al (2003). The secretory cells showed well developed granular endoplasmic reticulum and contained electronlucent secretory granules as mentioned by Chaudhry et al (1983) and Hakim et al (2002). These electron microscopic findings support the moderate alcianophilic and fuchsinophilic reaction (Pinkstaff,1975; El-Shafey et al, 1980; Pinkstaff and Salem and Bareedy,1987). Abuzaid et al (1990) stated that the positive alcianophilic reaction indicates mucous secretion. After birth, the secretory adenomeres increased greatly in size and number and the granular endoplasmic reticulum became extensive. The secretory granules increased greatly in number and size supporting the massive increase in alcianophilic reaction which reach its maximum at 4 months of age. These results are in agreement with those of Chaudhry et al (1983), Nagato et al (1998) and El-Bargeesy et al (2001).

The cells of the serous demilune contain oval to spherical basal nuclei and their cytoplasm was filled with rounded electron dense granules.Chaudhry et al (1983) and Hakim et al (2002) described these granules as serous granules. Our study revealed that these granules increased gradually in number after birth and showed weak alcianophilic reaction as mentioned by Nossir and El-Lakany (1988) and Abou-Easa (1993). As recorded in different animals by Borghese (1950), Yadm (1985), Nossir and El-Lakany (1988), Abou-Easa (1993), Nagato et al (1998) and Hakim et al (2002) our study revealed that the mandibular gland was formed mainly of mucous adenomeres with few serous demilunes.

The duct system of rabbit mandibular salivary gland was firstly recognized at the 20th day of fetal life as small intercalated ducts lined by simple cuboidal epithelium. These ducts appeared branched, increased in number and diameter at 22 days upto the fullterm but showing no signs of secretory activity. Similar results were reported by Yadm (1985), Latshow (1987), Abuzaid (1990) and El-Bargeesy (2001). After birth inter and intralobular ducts became numerous and well developed. Also striated ducts were clearly obvious and the luminal contents of all ducts showed moderate to strong alcianophilic reaction. This agree with the findings of Yadm (1985) and El-Bargeesy (2001).

The myoepithelial cells were firstly recognized at 29th day intrauterine as flattened elongated cells with poor cytoplasm. They were seen around some acini as mentioned by Szymanska (1963), Borghese et al (1974), Chaudhry et al (1983) and Sivakumar et al (2003). After birth, the myoepithelial cells were very clear encircling all acini either mucous or mixed in addition to the excretory ducts (Chaudhry et al, 1983; Hardy and Kramer, 1998 and Hakim, 2002). The latter author recorded that the myoepthelial cells have a contractile function as their cytoplasm contains myofibrils. The efficiency of their function was supported by the desmosomal junction between them as recorded in the present work .

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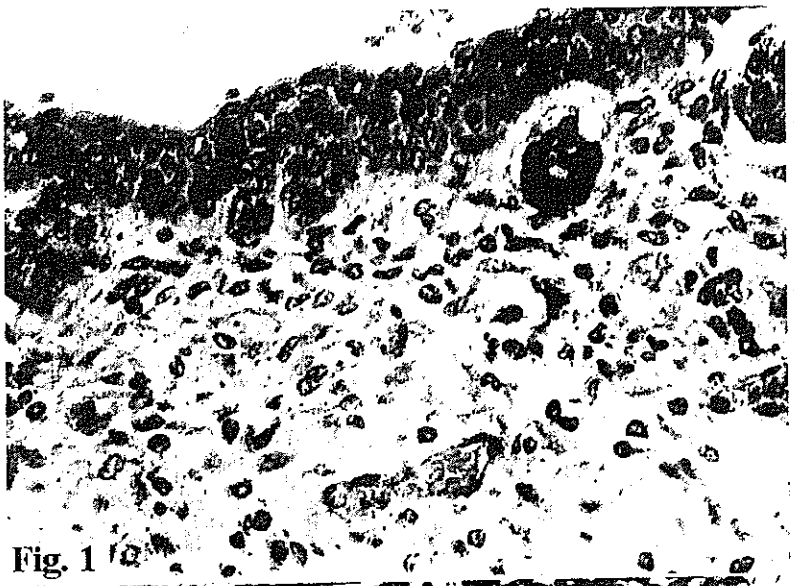

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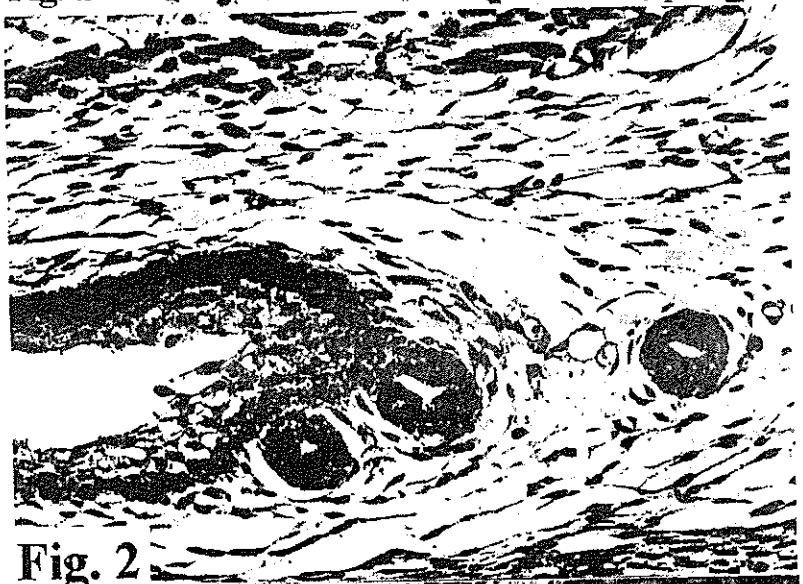

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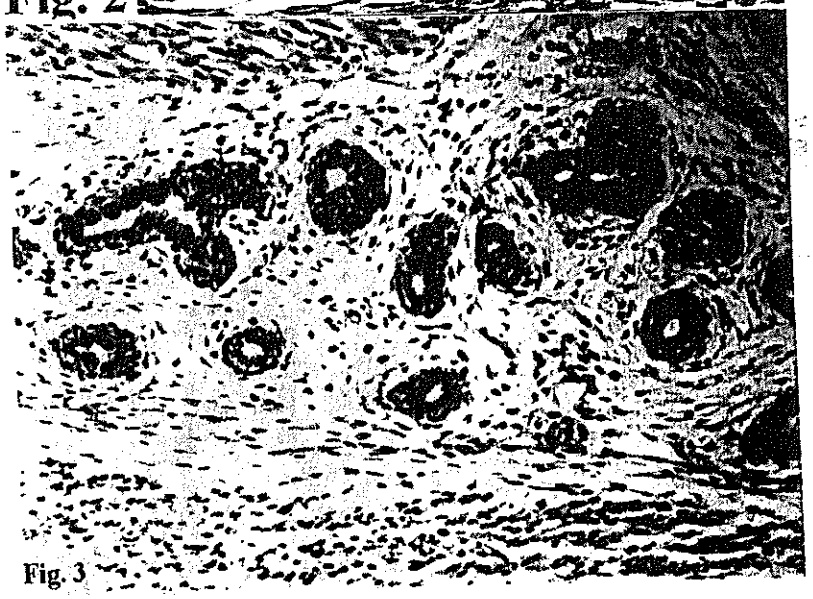

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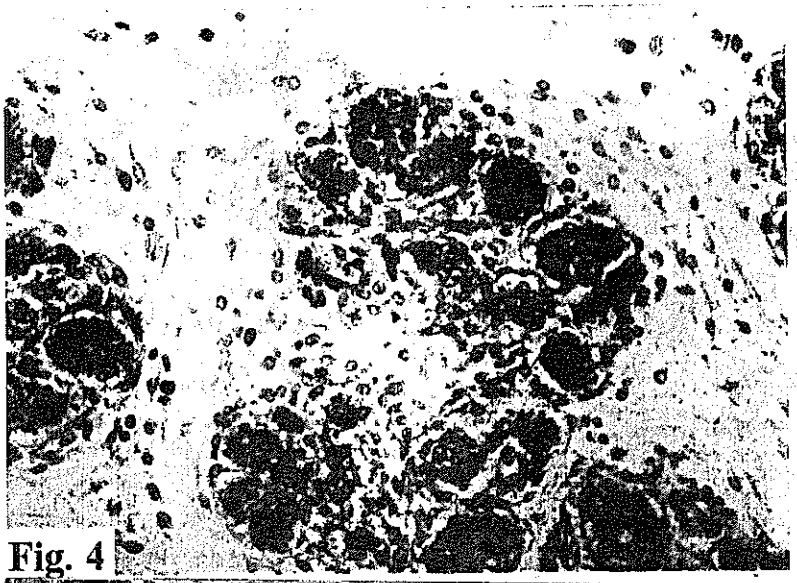

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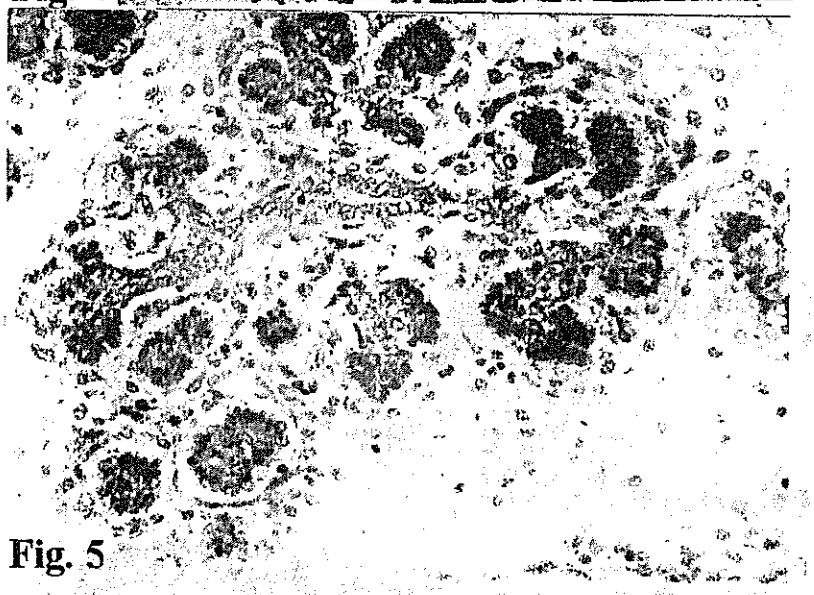

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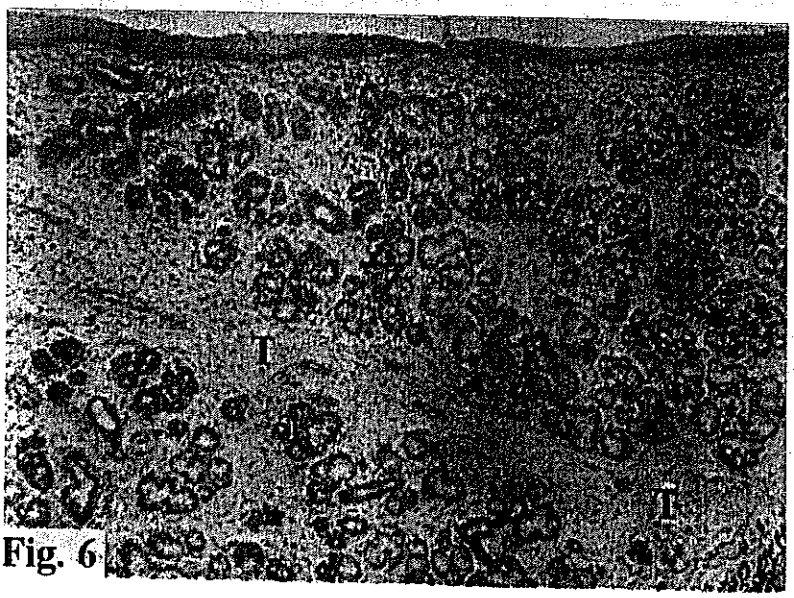

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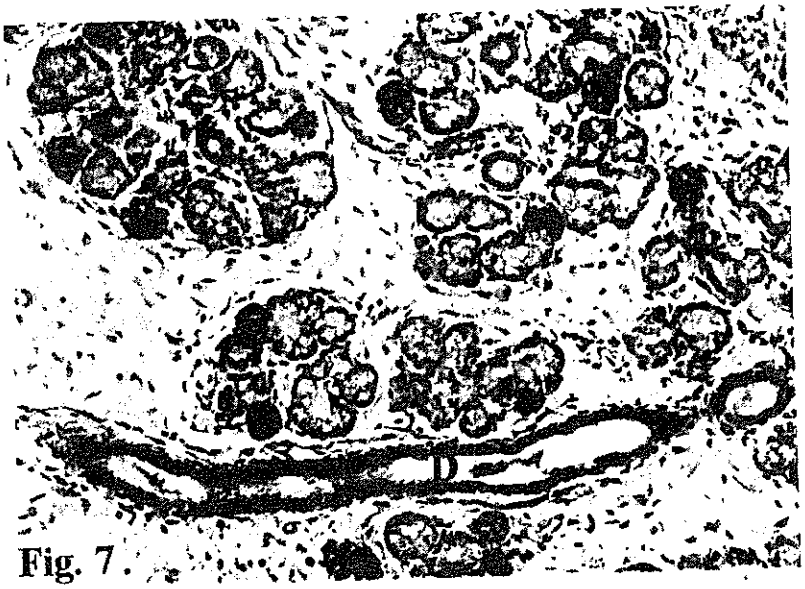

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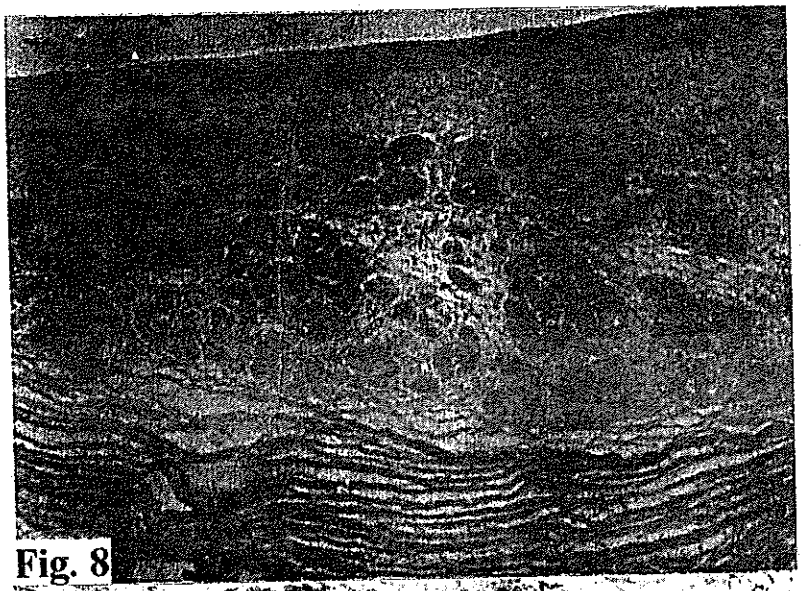

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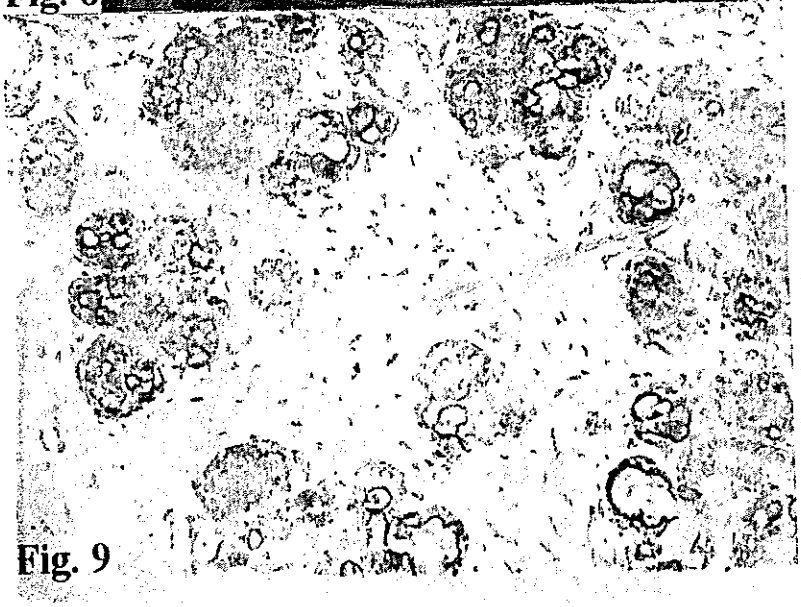

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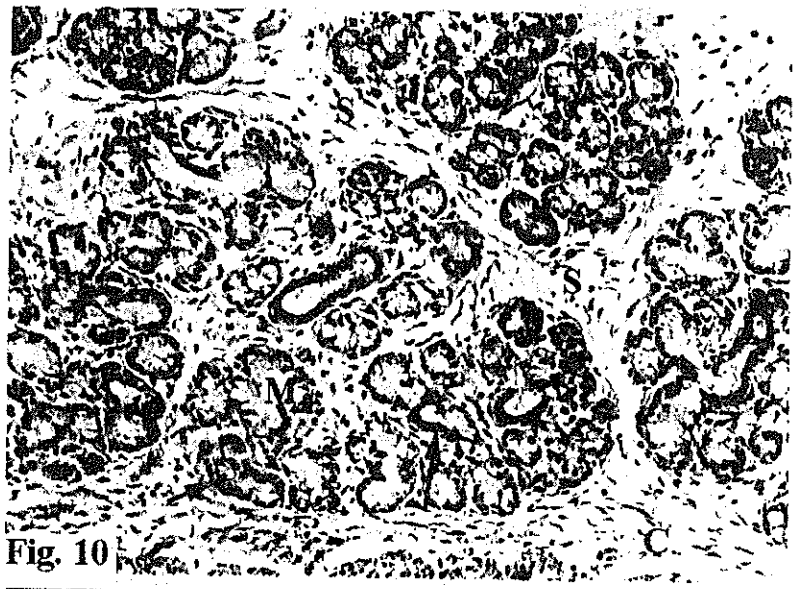

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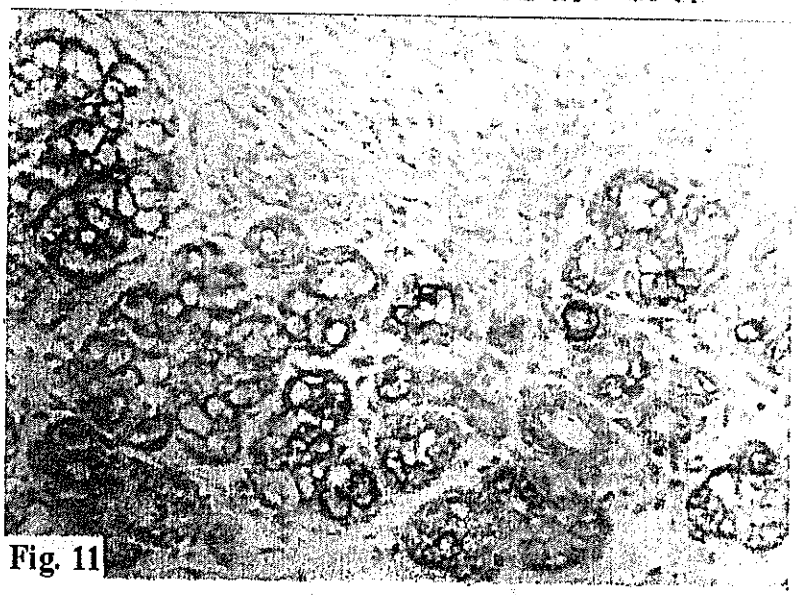

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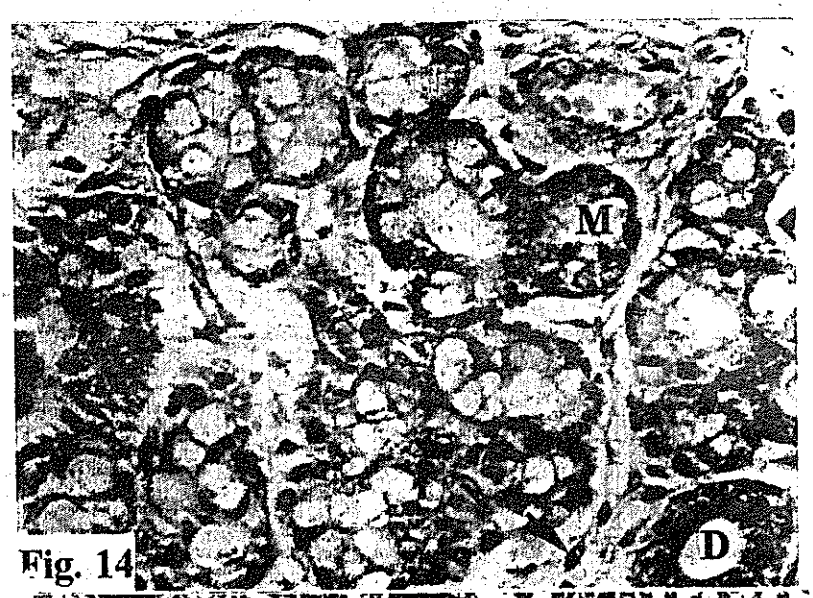

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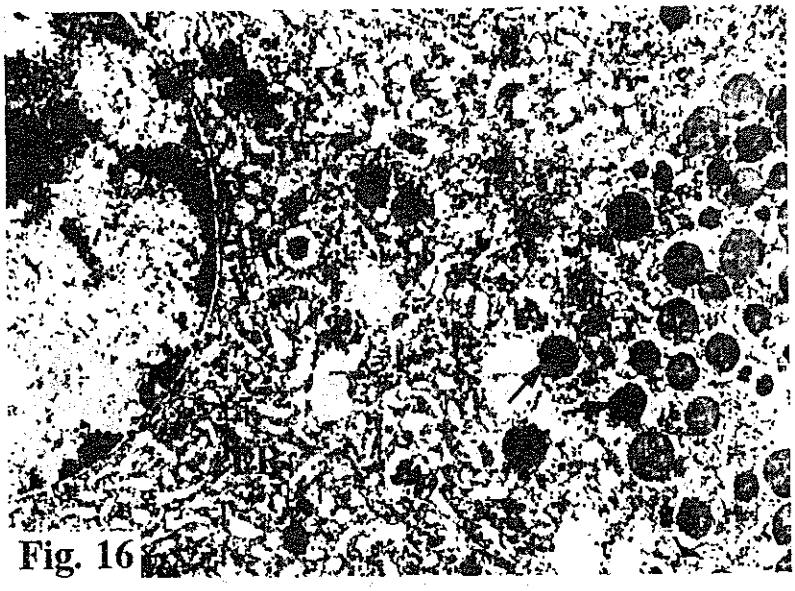

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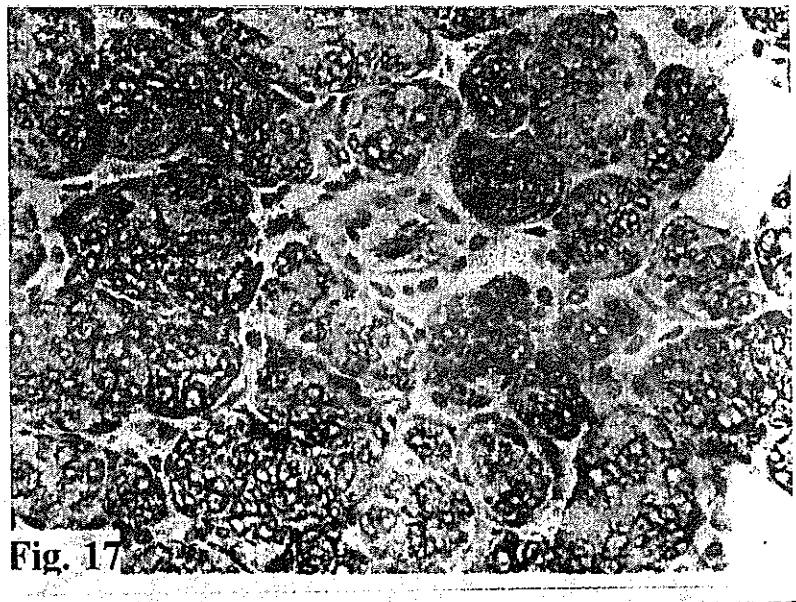

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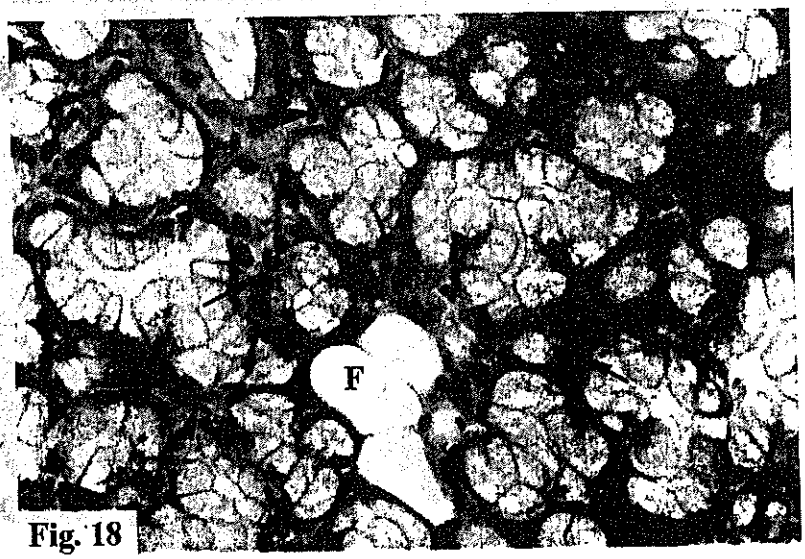

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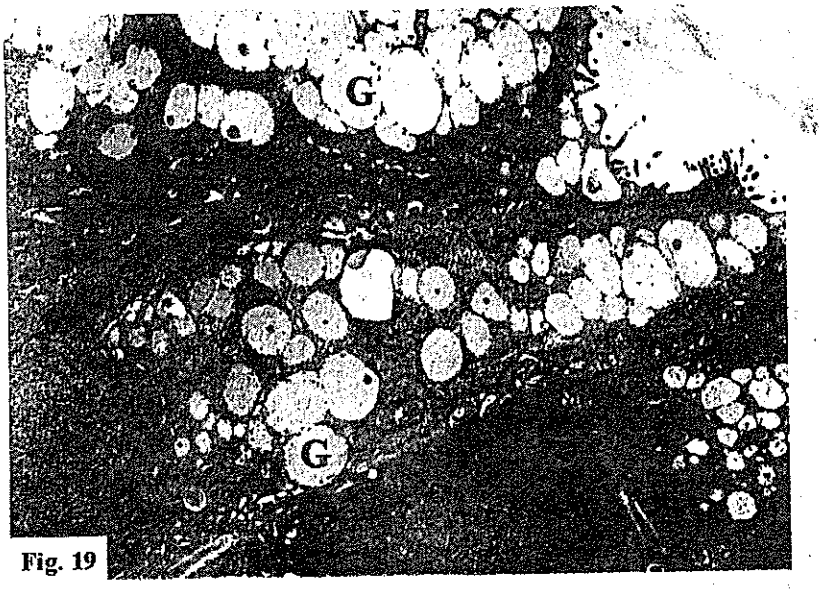

Fig. 19

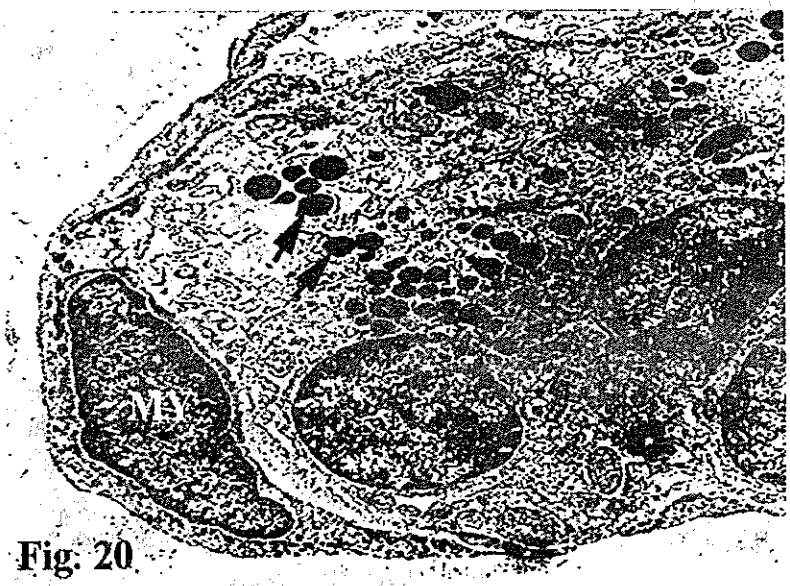

Fig. 20

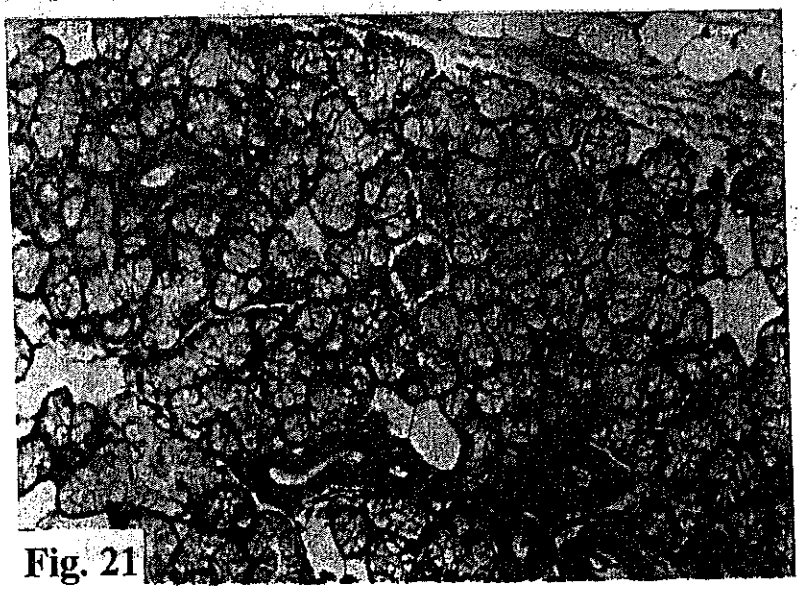

Fig. 21

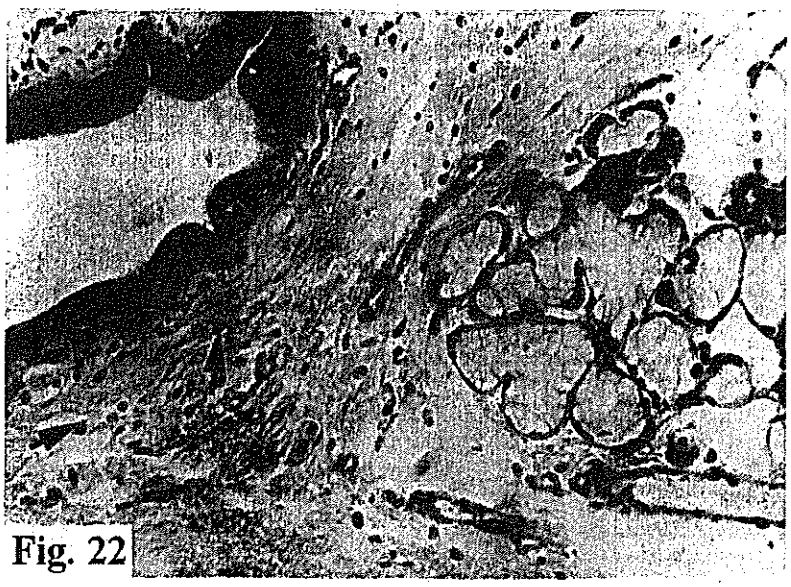

Fig. 22

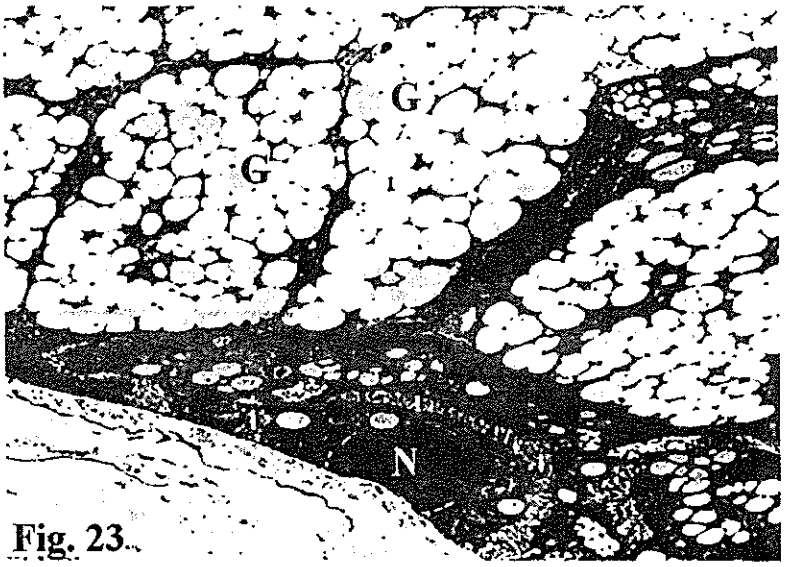

Fig. 23

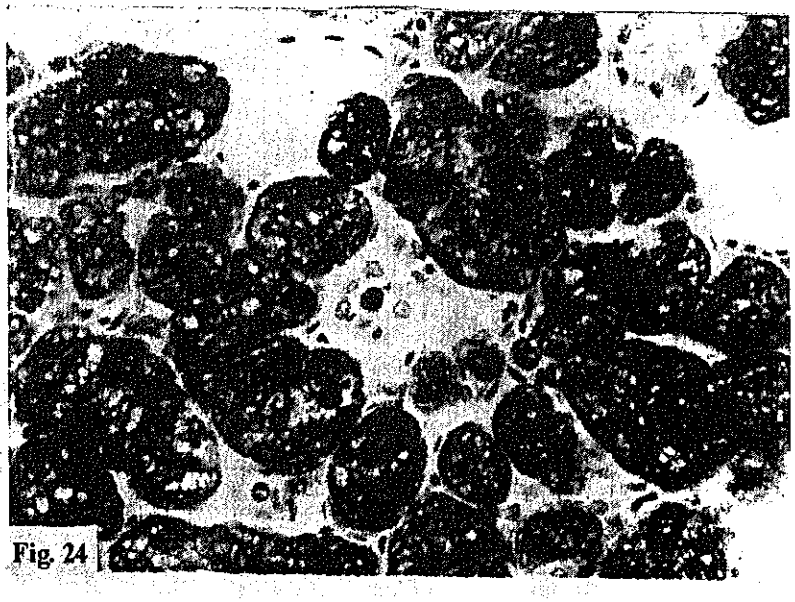

Fig. 24

الملخص العربي

التطور النسيجى للغدد اللعابية الفكية فى الأرانب النيوزيلاندية البيضاء

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قسم الخلايا و النسجة – كلية الطب البيطرى- جامعة بني سويف

أجريت هذه الدراسة علي الغدد اللعابية الفكية لعدد 20 من الأجنة تراوحت أعمارها بين 11-30يوما اضافة الي 15 عينة أخذت من أرانب تراوحت أعمارها بين 1-120 يومـــا. حفظت هذه العينات فى فورمالين 10% ثم قطعت و جهزت للفحص بـــالمجهر الضـــوئى و الألكتروني و أظهرت النتائج ما يلى:-

* بدأت الغدد اللعابية الفكية فى الظهور علي هيئة براعم طلائية مصمتة عند الأخـــدود اللساني اللثى في أجنة عمرها 13 يوم. بعد ذلك ظهرت تجاويف فى قلب هذه البراعم لتأخـــذ شكل أنيبيبات أو قنوات آخذه في التفرع.

* ابتدأ ظهور العنبات المخاطية و كذلك الهلالية المصلية في اليوم العشرون من الحمل ثم تزايد عددها و حجمها بعد ذلك حتى الولادة حيث اظهرت تفاعلا ايجابيا لصـــبغة الألســـيان الأزرق و الالدهيد فوكسين. كذلك ظهر منشأ القنوات المقحمة و ازدادت فى التفـــرع و قبـــل الولادة ابتدأ ظهور القنوات المخططة. كما لوحظ أيضا ظهور ألياف شبكية حول الفصيصات عند عمر 22 يوم من الحمل و كذلك ظهر الغطاء الليفي المحيط بالغدة و تزايد تدريجيا فـــي التطور حتى الولادة .

* بعد الولادة أصبحت الغدة محاطة بألياف غروية و شبكية تحتوى علي أوعية دموية و قنوات و امتدت هذه الألياف بين الفصيصات. و احتوت هذه الفصيصات علي وحدات افرازية مخاطية و كان القليل منها مغطى بالأهلة المصلية و أحيطت كل الوحدات الافرازية بخلايـــا طلائية عضلية. و وجدت خلايا دهنية بين الفصيصات و ازداد عددها مـــع تقـــدم العمـــر. و وصلت الغدة الي قمة تطورها النسيجى و الافرازى عند الشهر الرابع من العمر.