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A Comparative Study of the Salivary Glands of Certain Animals with Widely Diverse Food Habits

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A COMPARATIVE STUDY OF THE SALIVARY GLANDS OF CERTAIN ANIMALS WITH WIDELY DIVERSE FOOD HABITS

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I certify that I have read this thesis and that in my opinion it is fully adequate, in scope and quality, as a thesis for the degree of Master of Science.

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INTRODUCTION

The relationship of the morphology and physiology of the digestive system to the food habits of an animal has long been of interest to investigators. Frequent allusions to such a relationship are found in the literature, but specific information is not readily available. Most work of a comparative nature has been concerned with gross morphology on laboratory and domestic animals, and detailed histological studies on most groups of mammals are lacking.

Interest in this area was stimulated largely by the question of whether the morphology of mammalian salivary glands may be correlated with food habits, on the one hand, and with phylogenetic patterns on the other. Since food habits are ordinarily related to taxonomic groupings, correlation with food habits would also suggest correlation with the phylogenetic relationships. Although there are closely related animals with widely divergent dietary habits, such as the insectivorous and frugivorous bats, no such examples were available for this study, and there are no satisfactory accounts in the literature on which to base comparisons.

Available time and material restricts this study largely to a comparison of the gross weight and microscopic structure of the salivary glands in a series of six mammals ranging from the carnivorous bobcat to the seed eating kangaroo rat.

Since the salivary glands do not have identical physiological roles in the various groups of mammals, both the relative sizes and the details of structure might be expected to vary from group to group. Relative metabolic rates in small and large mammals, as well as the range of functions, must be considered in interpreting data.

A consideration of the eating habits of a strict carnivore which bolts its food suggests little need for the enzyme salivary amylase, but does indicate a use for mucin and serous fluids for lubrication and moistening the mouth cavity. For herbivores and omnivores the presence of a ferment such as amylase to initiate the breakdown of starches would be advantageous.

It is the purpose of this study to survey the literature concerning variation in salivary gland function and structure, and to study these glands histologically in a selected series of six mammals of differing food habits, and to show evidence of the variation one might expect to find.

GENERAL CONSIDERATIONS OF THE SALIVARY GLANDS

Superficially, the problem of salivary gland function appears to be a simple one. Much effort has been expended to determine the various aspects of function and morphology, but a review of the literature suggests that it has been difficult to arrive at definite conclusions, and reveals a confusing lack of agreement.

The main function of the salivary glands relates to feeding activities and is often manifested in interesting ways. For example: the toxic nature of the secretions of the modified salivary or poison glands of vipers is well known. Grundset (1958) refers to a substance in the saliva of the shrew which when injected into mice caused them to lose alertness, breathe heavily, become partially paralyzed, convulsed, and finally to die. The anteater is able to capture insects by means of sticky mucous secretions coating the tongue. It is of interest that the submaxillary gland of this animal is provided with a special receptacle (Andrew 1959) to hold the sticky mucous secretion, which, according to Dalquest and Werner (1952), has the consistency of Canada balsam.

Better known actions of saliva, which are associated more closely with the process of eating, include mechanical and physical effects that facilitate chewing, swallowing and tasting. In man speaking is facilitated by the moistening of mucous membranes.

It is usually considered that the principal chemical action of saliva upon food is due to salivary amylase, and that its presence is common among animals. Studies by Jung (1925), however, show the enzyme to be insignificant in the saliva of the dog, the ruminants and the horse; but is prominent in the pig, rabbit and man. It was found that in the

horse the saliva is much more active when recovered from the eosophagus. Sources of this increased activity are thought to be bacteria or glands farther back in the mouth and throat. Jung's conclusion was that, in animals without salivary amylase, the role of saliva is exclusively mechanical and physical. This conclusion cannot be substantiated, however, in view of further information.

According to Frischl and Kahn (1930), the removal of the parotid glands from the rabbit rendered the saliva almost inactive with respect to salivary amylase. A comparison of the amylolytic activity of both male and female rats and mice was made by Raynaud and Rebeyrotte (1950) under conditions of castration, adrenalectomy, and after injection of thyroid hormone. An atrophy of the tubular system of the submandibular gland appeared in the male mouse. This system, which is more extensively developed in the male mouse than in the female, is dependent for its maintenance upon androgenic hormones and thyroxine. The administration of the androgenic hormones to the female greatly augmented the secretion of salivary amylase, whereas the castrated male showed a marked decrease of secretion. The functions and morphology of the parotid glands were not affected. There is little difference between the tubular systems in male and female rats, and, weight for weight, the amylase activity is nearly equal. From the foregoing facts it seems probable that the tubular glands are responsible for the secretion of amylase by the submandibular glands of the mouse and rat.

There are additional properties of saliva which are not so well known. In man saliva has a buffering action which is dependent principally upon the bicarbonate ion. It was shown by Lilienthal (1955) that the extraction of bicarbonate and phosphate ions, while leaving mucin and proteins, removed saliva's buffering properties.

Babkin (1944) shows the importance of certain alkaline substances in the parotid gland of the sheep which, if lost through a permanent fistula, will reduce the animal to a state of emaciation and eventual death because of inability to neutralize the acids formed in large quantities in the rumen.

An interesting activity is claimed for the submaxillary and sublingual glands of albino rats by Screebny, Meyer and Bachem (1955). They state that "the submaxillary and sublingual glands of albino rats contain considerable amounts of an enzyme system which can act on casein, gelatin, and other proteins, but not on mucin or chondroitin sulfate, and therefore, appears to be a relatively nonspecific proteolytic system."

Cavazzuti (1948) reports a substance found in the saliva of the parotid gland which contains a mucinolytic substance which has the effect of decreasing the viscosity of the saliva of the submaxillary gland. Attention has been called by Pigman (1957) to enzymes capable of hydrolyzing hyaluronates found in the secretions of a variety of pathogenic organisms, in the venoms of snakes and insects, and in the saliva of leeches and certain insects.

An unusual use of salivary secretions is found in certain carnivores. Romer (1955) states, "in carnivores, generally, sweat glands are much reduced in number; the panting of a dog utilizes salivary evaporation on the tongue as a substitute." The gland or glands which supply the moisture is not revealed in this reference, but a probable source is the parotid gland.

Recent investigations by Shafer and Muhler (1956) give evidence for a function of the rat and human submaxillary glands as a "reverse thyroid gland." They have proposed that one of the important functions

is to control the concentration of thyroxine in the blood by deiodination and return of the iodide ion to the thyroid gland. Shafer and Muhler (1956) make reference to the work of Reiss, Halkerston and Badrick who found that the uptake of I^{131} by the salivary glands in some humans resulted in a salivary isotope concentration 600 times that in the blood. It appears that this concentrating activity of the salivary glands has some relation to the state of the thyroid function. Ryan and Kirkwood (1955) have shown that the administration to rats of thyroid blocking agents such as thiouracil or thiourea results in a marked atrophy of the submaxillary glands. They also quote the findings of Moehlig that obvious swelling of the salivary glands is often associated with hyperthyroidism. According to Ruegamer's (1955) experiments, on the other hand, the salivary glands of the dog were not active in the uptake of diiodotyrosine. He showed that the amount of iodine secreted in the saliva is directly related to the plasma level of iodide ion and is not influenced by thyroid hormone.

In addition to discussing the physical and chemical properties of saliva and salivary glands, statements are made linking size relationships of the glands to food habits. Andrew (1959) finds that the salivary glands vary somewhat in degree of development, in location, and in type of secretion according to the animal's way of life and the manner of feeding. Stormont (1928) notes that the submaxillary gland shows the greatest degree of variation when comparing different orders of animals, but that related animals within each order possess considerable similarity of morphological characteristics, although occasionally there are some individuals which do not resemble the group.

According to Andrew (1959) the parotid gland is larger than the submandibular gland in the fruit eating bats, but is smaller in the insectivorous types. He also states that the submandibular gland of the edentates is highly developed, but the parotid gland is relatively small. Dalquest and Werner (1952), however, are not in agreement concerning the identification of the larger gland, which they identify as the parotid.

Certain rodents whose diet is entirely from plant sources appear to have, relatively, the largest parotid glands. Andrew (1959) observes that the parotid glands of the beaver are exceptionally large, but that in the rat they are smaller, being about equal in size to the submaxillary glands. Although this statement may be true for the white rat, it does not represent the situation in Dipodomys agilis, in which the parotid glands are approximately four times the size of the submandibular glands (table 2). In referring to the ungulates, Andrew (1959) states that the parotid gland is almost four times the size of the submandibular gland. This is approximately correct for the horse; but the data in table 2 reveal important exceptions. According to Carmalt (1913a) only two of the three components of the typical mammalian alveolingual complement of glands are fully developed in the typical ungulates. These are the submandibular and the lesser sublingual glands. The greater sublingual gland is usually absent, but the lesser sublingual is usually greatly developed.

The salivary glands of the cetaceans and sirenians are said by Adams (1933) to have been reduced to vestiges because of their long association with water, but the glands of the Pinnipedia are not reduced in size compared to the condition in land animals.

Certain general histological considerations of the salivary glands are also of interest. It was Heidenhain who first coined the terms "mucous cells" and "albuminous cells" which are used so commonly (Pflüger 1870). Usually one considers that these terms apply to specific types of cells which can be differentiated by their staining properties. These terms are, however, general in meaning and loosely used. According to Stormont (1928), the determination of whether a cell is mucous or serous is only the first and a relatively unimportant step in gland analysis. Le Blond (1950) has coined a composite term "seromucoid" to describe certain cells which appear to stain as serous cells, but exhibit the histochemical properties of mucins. Thus it is difficult to attempt a comparative study on a purely morphological basis. A better comparison is possible using histochemical methods. This is well stated by Bensley (1908) who said, "no successful attempt has been made to subdivide the general classes of mucous and serous or albuminous glands into subordinate categories," and "any such subdivision must be on physiological or biochemical grounds, because of the lack of fundamental differences in the structure of the protoplasm of glandular cells."

MATERIALS AND METHODS

This project was begun by compiling a list of mammals which seemed desirable for this study because of their varying food habits. They were grouped as follows: carnivorous, herbivorous and omnivorous animals. Use was made of any mammal which became available within a given category. The animals collected included: the bobcat (Lynx rufus), coyote (Canis latrans), opossum (Didelphys virginianus), kangaroo rat (Dipodomys agilis and Dipodomys deserti), and goat (Capra aegagra). At a slaughter house glands were obtained from severed heads of the cow (Bos taurus).

For the purpose of this study the animals are arranged according to diet. The bobcat is more carnivorous in nature than the other animals within the group. Hamilton (1939) states that "our native cats are more strictly carnivorous than any of the meat eaters thus treated but they also make an occasional meal of fruits and berries."

Next to the bobcat, the coyote is the most carnivorous, obtaining the major part of its food from animal life, much of it in the form of carrion. Nevertheless, according to Hamilton (1939) "the coyote, like others of its tribe, feeds freely on whatever the country may offer," including vegetable foods such as fruit, cactus, juniper berries, rose seeds, currants, and others.

The opossum is an omnivorous creature. Hamilton (1939) has examined the stomach contents of several, finding such items as worms, snails, insects, snakes, young turtles, wild cherries, other fruits and berries. "Indeed," he says, "this species is probably as omnivorous as any flesh eater."

The remaining animals studied are strictly vegetarian but vary somewhat in preferences. Ordinarily the food of the goat consists of the leaves and twigs of shrubbery and trees. It will eat dried alfalfa hay, and enjoys grain mixtures which may be fed to it in captivity. The cow and horse are grazing animals, preferring fresh forage, but can subsist easily on dry foods. Both animals relish grains when available. Howell (1933) reports that the principal food of the kangaroo rat consists of grass seeds and root stocks of grasses, indicating a dry diet of a starchy nature.

The preparation of the animals for study consisted of determining and recording the weights in the cases when the complete animal was available. Average body weights were used for the cow. No horses could be obtained for dissection, but in the interest of further comparison data reported by Sisson and Grosman (1938) are included in table 2. As soon as possible after the death of each animal the glands were removed and weighed. Portions of each gland were placed in the following three solutions: neutral 10% formalin, Bouin's, and Zenker's fixatives. The tissues were then imbedded in tissuemat paraffin and cut at 7 microns thickness. Serial sections were made for routine staining, but single sections were chosen for special stains. The stains consisted of hemotoxylin and triosin, Krichesky's modification of Mallory's aniline blue collagen stain, periodic acid Schiff stain, toluidine blue, iron hemotoxylin and Mayer's mucicarmine. Sections from all tissue specimens received the routine stain, but only selected sections from each animal received the special stains. The slides were first examined for cytological differences among the various glands; the special stains were used primarily as simple histochemical tests in order to more accurately classify the secretory activities of the various cells.

Toluidine blue was used because of its metachromatic staining properties with sulfated mucopolysaccharides. At times, toluidine blue is erratic in staining and, in order to improve its action, it was processed according to the technique of Robinson and Bacsich (1958). In brief, this consisted of precipitating a filtered 1.0% solution of toluidine blue by adding a saturated solution of KI drop by drop, and washing the precipitate on a filter. The precipitate was dried, and a solution containing 0.3 gm. of precipitate per 100 ml. distilled water was used for staining.

Metachromasia was studied by Lison, who reported that the effect was due to the sulfate esters in high-molecular-weight carbohydrates (Pigman 1957). It has since been demonstrated, however, that metachromasia is not confined to sulfate esters, but is exhibited by the nucleic acids and a number of other substances (Pigman 1957). The so-called mucin stains do not necessarily reveal all the various mucopolysaccharides in tissues. They react particularly with the acid mucopolysaccharides.

The Schiff reaction was used for comparison with the results obtained by toluidine blue and mucicarmine because it shows "a much wider distribution of reactive material than any of the other methods since it stained all periodate oxidizable polysaccharides whether acid or neutral" (Gomori 1954). Le Blond (1950) stated that the interpretation of the results obtained by use of periodic acid-fuchsin-sulfurous acid (FSA) method required some caution. Studies showed that many substances other than carbohydrates could be oxidized to aldehydes by periodic acid, while some carbohydrates did not react to this acid. By a process of elimination, Le Blond (1950) was able to exclude from

consideration a number of carbohydrate-protein complexes which ordinarily give a mucin reaction. Some which are water soluble and fat soluble are removed by ordinary means of histological preparation. Other substances would be only slightly reactive due to the masking or substitution of the reactive glycol groups necessary for an intense reaction by this technique. Le Blond's conclusion was, therefore, that mucopolysaccharides were mainly responsible for the stain reactions in salivary glands. Of these, mucoitin sulfates and neutral mucopolysaccharides would be most abundant. Mucopolysaccharides, such as heparin, hyaluronic acid and chondroitin sulfate, were not expected to be present within the epithelial cells.

OBSERVATIONS AND DISCUSSION

The histological description of the various glands in this study include a discussion of those features which are peculiar to the individual gland. The stromal components are mentioned only when they are sufficiently different to be of interest.

Besides the usual three well-known salivary glands of mammals in general, a fourth is found in the orbit of some carnivores and is referred to as the orbital or infraorbital gland (Gland of Nuck). Hyman (1942) considers the parotids, submandibulars, sublinguals and infraorbitals to be the principal salivary glands of mammals. According to Blumenbach (1827) the infraorbital gland is found principally in the Canidae; however, it is listed by Reighard, Jennings and Elliott (1935) among the principal glands of the cat. In the present study it was found to be prominent in the bobcat and the coyote. Because it is considered to be a principal salivary gland, it is included in the histological descriptions and in the data of table 2 for both individuals.

Among the carnivores the sublingual gland presents variation in size between the divisions referred to as the major and minor sublingual glands. The minor sublingual gland may be extensive in some species or it may consist of only a few scattered elements in the vicinity of the lingual nerve in others (Carmalt 1913b). The major sublingual gland is the better developed of the two parts in the bobcat and coyote, and will be the portion described in this study.

Bobcat, Lynx rufus

Infraorbital gland: The infraorbital gland appears to be a pure mucous variety composed of exceptionally large acini which are

continuous with long duct-like corridors lined with mucous cells. Many branches arise from the main corridor and terminate as alveoli. The corridors, centrally, are continuous with excretory ducts having a similar or larger caliber (fig. 16). The lumens of both acini and corridors are large and distended with mucous. Dark staining cells are distributed in an irregular manner as a part of the cellular wall of the alveoli. At times they appear as demilunes and again as individual cells squeezed into a spear point between distended mucous cells. Although many such cells give the impression that they lie at the base of mucous cells, a close inspection reveals that the apex of each dark cell reaches the lumen of the acinus. Oblique or horizontal sectioning may cause such cells to seem larger. The cells just referred to are not to be confused with the basal cytoplasm of distended mucous cells which is also dark staining. Most of the dark cells are to be found in the distal ends of the gland elements rather than along the tubular corridors. The cells of the mucous type were heavily stained by all mucin stains (table 1).

Parotid gland: The majority of the cells and acini of the parotid gland are of the serous type judging by routine staining qualities. They are large compared to the general type of serous acini. The cells are tall but irregularly shaped, fitting together at many angles. Large refractile granules fill the cells, some cells being densely packed and others quite loosely arranged. When the tissue is stained routinely, the granules are colored orange-red. With Krichesky's modification trichrome they are deep blue. A small cell with densely packed granules is ordinarily more deeply stained, whereas greatly distended cells having loosely arranged granules are lighter in color. Apparently the granules undergo a change which eventually causes them to disappear. Stages can

be seen in which the granules are faintly stained while the cell is acquiring the characteristics of mucous cells. Mucous cells and acini are numerous and scattered at random throughout the parenchyma. The serous type cells appear as one sort and are considered by the writer to be sero-mucoid cells. These observations are reinforced by a consideration of the staining reactions of the parenchymal cells which will be noted later.

The lumen of the acinus is small (1 to 3 microns in diameter). Intercellular canaliculi have been observed to leave the central canal to penetrate between all cells of the alveolus. The canal of the alveolus is continuous with the lumen of a long intercalated duct. The arrangement of the acini and intercalated ducts is typical of the classical "bunch of grapes" and is referred to as a compound alveolar gland.

The staining characteristics of this gland are unique. A strong positive Schiff reaction was present in all cells of the parenchyma (fig. 5). The staining substance was in the form of granules in the dark cells. The reaction was stronger in the mucous cells. Metachromasia was moderately manifested in the granules of all dark cells with toluidine blue, but was stronger in the mucous cells. Mucicarmine stained only the mucous cells (table 1).

Submandibular gland: Superficially there seemed to be many dark staining serous cells interposed between mucous acini. Upon closer inspection, however, it is seen that these dark staining cells are in a variety of stages of activity. Many cells of an acinus are completely filled with mucous granules. Other cells, those that are of the dark variety, are usually partially filled with the same granules but may have few or no noticeable granules (fig. 27). In other words, the probability

is that all of the cells, both dark staining and mucous filled, are one type of cell but in different stages of activity. The dark staining cells are a part of the wall of the acinus with their apices extending to the lumen of the acinus (fig. 28). The individual cells of the demilune formations showed no characteristics which would suggest that they are different from dark cells in other positions. Demilunes are said to be present in the cat (Bensley 1908), but Heidenhain, as quoted by Pflüger (1870) states that "the submaxillary gland of the dog when the mucous is withdrawn from it, no longer presents the demilune, but resembles the same gland in the rabbit."

Intercellular canaliculi are thought to be characteristic of serous cells (Maximow and Bloom 1957). In this study, however, canaliculi were observed to be present between the partially filled mucous cells. No specific reference to the presence of canaliculi between mucous cells has been found in the literature. Pflüger (1870), however, refers to extremely fine tubuli being given off from the central canal of the acinus, but he does not specify the type of gland. The canaliculi could not be observed for certain between the fully distended mucous cells, but could be observed between the partially filled mucous cells.

Recent work by Scott (1959) with the electron microscope indicates the existence of canaliculi between mucous cells in the rat. These are not in the classical form of canaliculi but exist as flattened spaces between cells. Small cytological structures resembling microvilli project into these spaces. Also, within the spaces, are found small nerve terminals which evidently travel between the cells. No secretion has been noted within these spaces and no definite functions can yet be assigned to these spaces. Recent textbooks (Maximow and Bloom 1957; Jordan 1947) do not agree with the foregoing, stating that secretory

canaliculi are absent. However, certain animals in the present study show the presence of both canaliculi and intercellular spaces quite readily. It is possible that distension of the cells by accumulating granules partially or completely obscures the canaliculi, since at times portions of these channels are seen in spite of the irregular cell outlines. The canaliculi were always surrounded by an area of clear protoplasm, unoccupied by any of the dark red staining granules present in the cytoplasm of the partially filled dark cells (fig. 29).

In general the staining reaction of the submandibular gland was highly metachromatic with toluidine blue. Even the darkest cells, which had no noticeable mucous granules, showed patches of metachromasia. The Schiff reaction stained all cells deeply (fig. 7), while with mucicarmine the mucous-filled areas stained more lightly than the dark serous type cells and the basal borders of the mucous cells (table 1).

Sublingual gland: The mixed serous appearance of the sublingual gland may be misleading. The cells are usually tall columnar except at the rounded ends of the acini where they become pyramidal in shape. These cells accumulate a mucin reacting material in a theca at their apex. The dark basal portions of the cells are quite extensive so that a section through the periphery of acini presents a picture of serous type cells, but sections through the apices of cells reveal the accumulation of secretory material. In cells which seem to be at the beginning of a secretory cycle an accumulation of Schiff positive granules may be seen near the apical border of the nucleus corresponding to the location of the Golgi apparatus. The cytoplasm of the cell stains darkly, more red than violet. With Krichesky's modification trichrome, the cytoplasm appears bright red due to many fine granules in the cytoplasm.

The duct system of the sublingual gland is very similar to that of the infraorbital gland, containing highly branching and rebranching corridors lined with secretory cells. The corridors empty into and are continuous with ducts of equal or larger size. The excretory ducts are lined with an overlapping type of flattened cuboidal epithelium.

Coyote, Canis latrans

Orbital gland: The orbital gland of the coyote is similar to that of the bobcat. A minor difference is noted in the excretory duct system which is slightly more extensive in the coyote. Branches from interlobular ducts enter the lobules. These intralobular ducts are not intercalated ducts, however, for this gland lacks a striated duct system. Intralobular ducts, which are few in number, gradually increase in size as they combine with others before reaching the interlobular duct.

Parotid gland: Although mucous acini have been noted in the parotid gland of the dog, none were observed within the parotid of the coyote. The parenchymal cells were of one type which are generally classed as serous cells. They are irregularly pyramidal in shape and are arranged in acini, containing few cells. The compound tubulo-alveolar system might preferably be described as a compound tubular gland because a prominent alveolus is not present. Most cells are completely filled with bright orange granules when colored by the routine stain. The nucleus was not displaced or distorted by the accumulated granules. Small cells were more heavily stained and the granules were more tightly packed, but the large cells were more lightly stained due to the loose arrangement of the granules. No discrete granules could be observed in the lumen of the duct system, although an amorphous secretion of the same

color was present. The granules were not observed to become faint and disappear, neither was a mode of secretion apparent. An intercellular canalicular mechanism could be seen, but, as stated previously, its function is not understood. A few individual cells containing a clear nongranular cytoplasm were observed among the granular cells, but they did not resemble mucous cells.

A strong reaction with the Schiff stain was significant. Variations of intensity indicated different stages of cellular activity. Other mucous stains failed to give a mucin reaction with the cells (fig. 4).

Submandibular gland: The secretory cells of this gland in the coyote seem to be two different types. This has been the basis of a controversy since Heidenhain described the demilunes which he called albuminous cells. Stormont (1928) referred to the demilunes of the cat and dog as special serous cells which would take the place of the older cells that were undergoing disintegration. Pflüger (1870) felt that the appearance of the demilune was a fixation artifact caused by the pressure of the mucous upon the cytoplasm basally. He states that in the fresh condition he was unable to observe the demilune. It is true that the granules compress the cytoplasm of mucous cells, but in the present study it was observed that the nuclei of the demilune cells are slightly oval and vesicular, whereas, the nuclei of the mucous cells are usually compressed and pyknotic. The demilune always appears to occupy a unique position in relation to the acinus. A small canal passes through the center of each acinus and branches into each outpocketing of the irregularly shaped acinus. The demilune caps the end of the canal which flares out beneath (fig. 23). From the space below each demilune, intercellular

canaliculi diverge like the points of a star and extend between the cells almost to the basement membrane. The canaliculi have been observed by Pflüger (1870).

The lumen of the acinus is continuous with the canal of a small intercalated duct which serves a group of acini. The intercalated duct in turn connects to a striated duct. Usually, only one intercalated duct is connected to any one branch of a striated duct, which is little more than a short stub from the main duct.

The mucous cells are deeply colored by the Schiff reagent (fig. 6) and the mucin stains, but the demilune cells and the cytoplasmic border of the mucous cells fail to give a positive test for mucin with toluidine blue and Schiff's stain. Mucicarmine stains the cells of the demilune and the mucous cells, but the duct system does not react with any of the foregoing stains (table 1).

Sublingual gland: No differences were noted in epithelium or in staining reactions of this gland compared to the sublingual gland of the bobcat.

Opossum, Didelphys virginianus

A study of the morphology of the salivary glands of the opossum reveals some unique features. Carmalt (1913a) states "this marsupial presents an extremely simple and generalized type of mammalian salivary organization approaching rather closely the conditions encountered in the average arctoid carnivore." This statement apparently refers mainly to the gross organization.

Parotid gland: The parotid gland of the opossum is a compound tubulo-alveolar type, the alveolus of which is composed of a closely packed

group of irregularly shaped polyhedral cells. The nucleus is large compared to the size of the cell; it is round or oval and highly vesicular. The cells of the alveolus are of various sizes and heights so that their arrangement in the acinus does not leave room for a central canal. Each cell appears to be surrounded by a space (fig. 30) which is part of a network draining the secretion toward the tubular portion of the gland. The tubular portion gradually acquires a central canal as the cells slowly decrease in size but remain cuboidal in shape, while at the same time the lumen increases in size until, as an intercalated duct, it meets and merges into a striated duct.

The staining reactions were almost negative with the mucin stains and Schiff reaction (fig. 2). The secretion within the ducts gave a positive Schiff stain. It appeared in some areas as if the ducts themselves had contributed the secretion (table 1).

Submandibular gland: The parenchyma of the submandibular gland is arranged in the form of compound tubulo-alveolar glands. The tubular portion of the gland is connected to a striated duct by an intercalated duct which is shorter than that of the parotid. The striated duct system is unique in that the individual branches travel together as a bundle in the middle of a lobule rather than scattering generally through the parenchymal cells (fig. 22).

The glandular cells appear to be of two types. It cannot be definitely known whether they are different cells or merely represent different activity phases. The general arrangement and form of the acinar cells is similar to that of the parotid. Although the majority of the cells of the submandibular gland contain mucous granules, many dark staining cells can be found located principally in the peripheral parts

of the acinus. The acinus and tubular portion each contains a central canal which is bordered by the apices of all cells both dark and light (fig. 30). Both types of cells retain the canalicular spaces as described in the parotid gland.

The cells filled with mucous granules became heavily stained by the Schiff reagent, whereas the dark cells failed to react (fig. 9). Mucicarmine stained the dark cells heavier than the mucous cells, but in both cases it was weak. Toluidine blue produced no metachromasia in any part of the gland (table 1).

Sublingual gland: The sublingual gland of the opossum resembles the infraorbital gland of the carnivores. Here is found a highly branched system of extensive corridors lined by a tall variety of columnar cells appearing to be a mucous type. As the branches move away from the main corridor, the cells are reduced in height finally resembling those of a normal mucous acinus. On these peripheral branches a dark border of cells can be seen. These appear to be separate cells, for the compressed nucleus of the mucous cell may often be found just within the dark cell with its nucleus.

No ordinary duct system is seen within the area of the parenchymal cells. The long corridors serve as ducts until the periphery of the lobule is reached, where it joins a duct containing cells of reduced height no longer resembling mucous type cells. The outer diameter of this duct is as great as that of the corridor continuous with it.

The Schiff reaction is intense within the mucous type cells (fig. 14); however, the dark border cells and the condensed cytoplasm of the mucous cells do not stain. Metachromasia with toluidine blue is weak and erratic. At times all cells of an acinus will stain moderately, and

at others only a cell here and there among nonstaining cells will exhibit the reaction. Mucicarmine behaved similarly to toluidine blue (table 1).

Goat, Capra aegagra

The principal salivary glands of the ruminants consist of the parotid, submandibular and minor sublingual glands. The minor sublingual glands, which occupy the greater part of the length of the alveolingual gutter, develop into a prominent gland, but the major sublingual glands do not become significant. The minor sublingual glands are those described under the heading of sublingual glands of the ruminants.

Parotid gland: A number of observations contribute to the impression that the parotid gland is very active. The nuclei which are highly vesicular, fill a major portion of the cell, for the cells are not large compared to other parotid glands. The cytoplasm, rather than being filled with granules, has a foamy texture. The cells are arranged about a large central lumen of an irregular shape due to the saw-toothed arrangement of the triangular, ragged apices of the cells. The rough cell borders were apparently due to secretion blebs. The cells often are semi-isolated from each other by deep lateral clefts which penetrate nearly to the base of the cell.

The large lumen of an intercalated duct, bordered by a flattened epithelium, empties into striated ducts with exceptionally large lumens, the purpose of which seems to be to transmit large volumes of secretion. Another evidence of high secretory activity is the presence of a rich blood supply of large vessels accompanying the divisions of the duct system, and an extensive capillary network.

None of the mucin stains reacted with the parenchymal cells or the duct system, except for goblet cells which become numerous in the major excretory ducts (table 1; fig. 1).

Submandibular gland: The submandibular gland in popular terminology is a mixed mucous gland consisting of mucous acini and the ever present demilunes. The demilunes are not to be thought of as accessory or as appendages adhering to the bases of the mucous cells. They consist of dark staining cells, each composing a segment of the alveolar wall, and each having access to the lumen by a pyramidal shaped apex.

Each dark cell is filled with bright red refractive globular granules when stained by Krichesky's modification trichrome. The granules have a dark border and a lightly stained center. This is due either to the refraction of light or to a peripheral layer which stains while the center does not. In the smaller cells the granules are densely packed. In larger cells the apex becomes loosely filled with granules separated by many clear spaces; at times the apex appeared as a clear vesicular space. It is not possible to determine if this represents a phase of mucous cell formation, or if the cells are of a special type, for the fate of the granules was not apparent. No discrete granules could be found in the lumen of the acinus.

The stain reactions for all cells are positive with the Schiff reagent (fig. 10). The granular cells present a more homogenous texture than the ropy coagulated secretion product of the mucous cells. The ducts gave a faint reaction in the apical portion of the cell. The mucous cells stained metachromatically, but the granular cells stained

only with the basic blue color of the dye. Mucicarmine stained both varieties of cells (table 1).

Sublingual gland: The sublingual gland also presents the aspects of a mixed mucous gland. The proportion of mucous type cells to dark cells is greater than the ratio in the submandibular gland and the demilunes are not as prominent and large. The general arrangement of the acini and the duct system is typical, but the striated ducts and the excretory ducts have a lumen of large diameter comparable to the duct system of the parotid.

A study of the fine structure of the cells reveals the presence of many stages of cell activity. Dark staining cells often exhibit a theca of accumulated secretion at the apex, which varies in extent from cell to cell. The dark cells do not contain large globular refractile granules such as are found in the submandibular gland, but contain an irregular, basophilic granular cytoplasm. The cytoplasm is pushed towards the base by the accumulating mucous secretion.

The stain reactions of the Schiff reagent illustrate the fact that the cells are in a variety of stages of activity (table 1). Within an acinus, and even within a cell, differences of color intensity may be seen (fig. 18). The mucous filled portion of each cell is highly metachromatic, but the effect is not present in the dark cytoplasm. Mucicarmine reacted with the same cellular elements.

Cow, Bos taurus

The salivary glands of the cow are similar to the glands of the goat in both morphology and staining reactions. The glands will not be discussed in more detail except to note two further observations. The

first is the presence in the parotid gland of the richest blood supply yet noted. The color of the gland in the fresh state was of a darker red-brown shade than that of the other parotid glands, a fact which may be due to the better blood supply.

The second observation concerns the phase activities of the granule cells of the submandibular gland. Previously, in the goat, the apical portions of the cells showed the accumulation of more space and a lessening of the concentration of granules, but no further transitional stages could be noted. The granule cells of the cow presented all stages of activity in which mucous accumulations and scattered granules could be observed in the same cell. The granules become fewer and more weakly stained, suggesting that a cell fully acquires the characteristics of a mucous cell when the stain no longer reveals the presence of fuchsinophil granules (figs. 20, 21).

Kangaroo Rat, Dipodomys agilis

The three main glands are also found in the kangaroo rat. It is possible to remove all three in one group. The submaxillary glands are in contact with each other at the mid-ventral line of the neck region (Midgley 1938). The sublingual glands are somewhat lateral but partly overlapped by the submaxillary glands. Still more laterad are found the parotid glands. Midgley (1938) reported the presence of molar glands and thought they might be accessory parotid glands, although he did not make slides of the gland. Sections made of these glands in this study were found to be sebaceous in nature.

Parotid gland: The parotid gland of the kangaroo rat resembles pancreas, not only in arrangement of cells, but also in staining habit.

Two zones of color are formed within a cell, a basophilic base and an acidophilic apex. Tissue removed from one animal revealed small pyramidal, basophilic cells containing large centrally placed vesicular nuclei while parotid gland tissue removed from another animal of the same species showed cells that were lighter staining (more acidophilic), larger and more distended with secretion (figs. 31, 32). The nuclei which were irregularly shaped and less vesicular, instead of being located near the centers of the cells, were now pushed to the bases. The cytoplasmic contents had the appearance of mucin granules causing the cells to resemble mucous cells. The Schiff reagent produced a light reaction within the cells in one instance, but the other mucin stains gave no reaction (fig. 3).

Submandibular gland: The submandibular gland is, as has been stated before, the most variable of all the glands. This gland in Dipodomys is very unusual in appearance due to the great abundance of large ducts referred to by Le Blond (1950) as serous tubules. The submandibular glands of the white rat are similar in appearance, but those of the kangaroo rat contain larger serous tubules having a higher columnar epithelium (fig. 25). They appear to occupy between 40 and 50 percent of the area of any field. Boerner-Patzelt (1956), in a study of the salivary glands of the golden hamster, noted that the large ducts came to an abrupt ending. The duct system as found in the kangaroo rat was highly branched, each terminal branch ending in a rounded dome of undiminished size apparently ending blindly. Pflüger (1870) spoke of these ducts in the white rat as salivary tubes. He could not find any means of communication between the alveoli and these ducts. Le Blond (1950) also mentions the white rat but states that the acini contain atypical

mucous cells which open into the serous tubules or ducts through short intercalated ducts. After a great deal of searching, the intercalated ducts were found in the submandibular gland of the kangaroo rat. They are very fine and are lined by a low cuboidal epithelium. Although they collect from a number of acini, only one intercalated duct enters any one blind ending branch of a serous tubule. It enters somewhat obliquely, usually a short distance below the rounded end. Flattened epithelial cells enter between and line the tall columnar epithelium of the serous tubule, appearing like a nozzle projecting inside (figs. 24, 26).

The parenchymal cells of the submandibular gland are so similar to those of the parotid gland both as to morphology and staining reaction, that one description is sufficient. The presence of vacuoles was noted within large numbers of parenchymal cells of the submandibular gland, but not within those of the parotid. Often the globular spaces appeared similar to the fat droplets in the acinar cells of the mammary gland. Where vacuoles were present the nucleus was pushed basally and was cupped to conform to the shape of the spherical structure. The structures were noted in the submandibular glands of some kangaroo rats but not in others. They appeared in tissue preserved with fixatives containing formalin, but were not noticed in Zenker fixed material. Fresh frozen tissues were stained with Sudan IV for fat, but the results were negative. Boerner-Patzelt (1956) noted vacuolated spaces in the cells of the same gland in the golden hamster. He was unable to stain them and so could not determine their nature. Babkin (1944) has observed the appearance of vacuoles in the demilune cells of the dog when there has been overstimulation of the sympathetic nerves, but the nature of the secretion was not determined. It may be possible that in sacrificing the animal, an overstimulation of the sympathetic system produced this effect.

The acinar cells have been referred to previously as atypical mucous cells because in some distantly related animals with similar type glands, mucin reactions have been noted. Cohoe (1907) reported such a reaction for the hedgehog and for the rabbit.

The stain reaction for the kangaroo rat showed many Schiff positive granules in the acinar cells which were not so strong as to obscure the cell. The reaction, however, was intense within the cells of the serous tubules (fig. 11). Toluidine blue had no reaction and mucicarmine stained the elements only slightly.

Sublingual gland: The majority of cells in the sublingual gland are the mucous type, but dark serous type cells are present. In comparing the same glands from two animals of the same species, variations were present in the numbers of mucous and serous types. The activity of one gland was such that few dark cells could be seen, whereas the activity of the other gland revealed them in greater numbers. The dark cells were often in the form of a demilune in the end regions of the acinus, the apex of each cell bordering the lumen.

The mucous cells react strongly with the mucin stains and Schiff's reagent (fig. 11).

SUMMARY AND CONCLUSIONS

It is reasonable to assume that the size and function of the salivary glands are suited to the physiological needs of the animal. Thus, if the functions of the salivary glands were the same for all mammals, one might expect to find linear relationships revealed by the foregoing data. This is borne out in table 2 and 3 but with modifications.

In table 2 the animals are arranged according to size from smallest to largest. Total gland percentages are recorded in the first column and indicate an irregular inverse ratio of total gland weight to body weight. Romer (1955) calls attention to surface-volume relations as an explanation for the fact that in a group of large and small animals there is a notable difference in the relative sizes of the organs. Areas are proportional to the square of linear dimensions, whereas volumes are proportional to the cube of linear dimensions. When comparisons are made on the basis of weight, however, a rough volumetric relationship is determined. Romer (1955) also states that "the amount of food which an active animal needs is roughly proportionate to its volume." He places emphasis on the word "active" and emphasizes that it is not basal metabolism which is referred to, but the metabolism of the animal in its active state. Smaller animals generally have a higher rate of metabolism than larger animals and their activity is often related to metabolic rate.

An additional factor regulating food requirements is the Bergmann principle which states: "Under identical conditions all homoiothermal animals give off equal amounts of heat per unit of surface" (Hesse, Allee and Schmidt 1937). The major part of the food eaten is transformed into body heat. If body surface area is small compared to

body mass, the amount of heat radiated is less than that from a large surface area. Thus large animals exposing relatively less surface area may have an advantage over small animals. By this reasoning, the food needs of the smaller animals are proportionately greater than those of larger animals; needs which in turn may be expected to require proportionately larger amounts of saliva from proportionately larger glands. It is probable that the correlation of gland size to body weight shown in table 2 is influenced by metabolic rate and surface-volume relationships. Although the data for the bobcat and the coyote fall into proper sequence, the percentages are lower than would be expected in a linear relationship perhaps in part because these animals bolt their food and thus have no need for large amounts of secretion. Observations made on the parotid glands of the cow and goat indicate that the physiological activity of salivary glands may affect the rate of secretion, thus offsetting differences in size.

In table 3 the animals are arranged according to diet, and some correlations of the data can be made. The relative sizes of the parotid glands shown in the second column are probably the most significant. Large differences occur between the figures for carnivores and those for kangaroo rat and horse. This would be expected, but it is difficult to account for the relatively smaller size in the cow and goat as compared with the horse and kangaroo rat, since the diets, particularly of the horse and the cow, are not significantly different. The manner of preparation of food for absorption by the ruminants can account for some of the variation. In the coyote the relative size of the parotid gland is greater than that of the cow or goat, probably related, as previously suggested, to panting as a cooling measure in the dog family.

It is apparent, therefore, that the parotid gland variations among the animals studied do not show a linear relationship to diet but parallel more closely the total food habits which often depend upon other physiological factors of an individual nature.

A survey, through stain reactions, of all the glands of each carnivore reveals that the cells are of a mucous variety or give a positive mucin test to a greater degree than those of each herbivore. The highly mucous infraorbital glands of the bobcat and coyote augmented by the mucin reaction of the cells of the parotid glands indicates the mucoid character of the secretions. The findings for the opossum are not significantly different from those for the goat and cow. If differences in mucin production are present among these animals, they would be due to differences in secretory rates of the cells and to differences in chemical composition of mucins. The glands of the kangaroo rat are the least mucous in nature. The sublingual and submandibular glands secrete mucous but are comparatively small. The submandibular gland secretes a mucopolysaccharide principally from the tubular system, though some is formed in the acinar cells. Variations in mucin production are greatest between the high level of the purely carnivorous bobcat and the low level of the purely herbivorous kangaroo rat.

On the basis of glandular morphology, there is in this study greater similarity between the animals with close phylogenetic relationship than between the animals with similar diets. Nevertheless, there are morphological differences between the glands of the bobcat and coyote although they are closely related and have only slight differences in food habits. The glands of the opossum show still greater differences when compared with those of carnivores, but the food habits of the opossum are more varied, and its relationship to the carnivores is somewhat distant.

The glands of the opossum, however, show greater similarity to those of the carnivores than to those of the herbivores. The glands and the diet of the goat and cow are almost identical, and their phylogenetic relationship is close. The greatest differences can be observed between the glands of the cow or goat and the kangaroo rat, all of which have herbivorous diets but distant phylogenetic relationships.

Since many physiological functions probably influence the size and morphology of the salivary glands, it would be necessary to consider the animal as a whole in a complete correlative investigation.

During the course of this study some general observations of interest were made. The presence of intercellular canaliculi among all cells in the salivary glands of this animal group suggests that such structures may be a universal feature of the cells of salivary gland parenchyma. The presence of canaliculi has long been reported among what are usually referred to as serous cells. Serous type cells were found within acini of all the glands studied, including the highly mucous infraorbital glands of the coyote and bobcat. In some serous type glands mucous cells are not present; in others they are a regular feature. It may be that the basic cell type of all salivary glands is the "serous" type cell. In glands which normally exhibit mucous cells, it is possible that changes may occur under proper stimulation which cause the serous type cells to become mucous cells as needed. Evidence for this statement is based on: (1) the observed transitions between the serous and the mucous cell types, (2) the presence of canaliculi which persist between the transitional and mucous type cells, and (3) the stain reactions which reveal the presence of mucins in serous type cells, this having been noted in the parotid gland of the bobcat.

The question of the probable function of canaliculi between mucous cells is of special interest. If the canaliculi are functional, it is possible that the mucous cells can produce more than one type of secretion. The serous type cell may have the ability in some glands to form serous and mucous products as needed according to the stimulus received. Confirmation of this possibility must await the availability of more detailed information concerning the control of salivary secretion.

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TABLES

ILLUSTRATIONS

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TABLE 1
MUCIN STAIN REACTIONS

ANIMAL	SCHIFF			TOLUIDINE BLUE			MUCICARMINE		
	Acinar Cells	Demilune Cells	Ducts	Acinar Cells	Demilune Cells	Ducts	Acinar Cells	Demilune Cells	Ducts
Canis latrans									
Parotid	+++		0 - +++	0		0	+		0
Submaxillary	++++	+	0	++++	0	0	++++	++++	0
Sublingual	++++	0	0	0	0	0	+	0	0
Orbital	++++	++++	+++	++++	0	0	++++	++	0
Lynx rufus									
Mucous	++++		0	++++		0	++++		0
Parotid Serous	+++		0	++		0	0		0
Submaxillary	++++	++++	0	++++	++++	0	++++	++++	0
Sublingual	+ - ++++	0	0 - +++	0 - ++++	0	0	0 - ++++	0	0
Orbital	++++	0	0	++++	0	0	++++	0	0
Didelphys virginianus									
Parotid	0		+	0		0	0		0
Submaxillary	++++	0	0	0	0	0	+	+	0
Sublingual	++++	0	+	0 - +++	0	0	0 - +++	0	0
Dipodmys agilis									
Parotid	+		0	0		0	0		0
Submaxillary	++	0	++++	0	0	0	0	0	0
Sublingual	++++	0	0	++++	++++	0	++++	0	0
Capra aegagra									
Parotid	0		0	0		0	0		0
Submaxillary	++++	0 - ++++	+	++++	0	0	++++	++++	0
Sublingual	+ - ++++	0 - ++++	0	++++	0	0	++++	++++	0
Bos taurus									
Parotid	0		+	0		0	0		0
Submaxillary	++++	++++	+	++++	0	0	++++	++++	0
Sublingual	++++	0 - ++++	0	++++	0	0	++++	0 - ++++	0

TABLE 2

RELATIVE GLAND SIZES, BY ANIMAL SIZE

Animal	Total Gland %	Parotid	Submaxillary	Sublingual	Infraorbital
Dipodomys agilis	.67	76.0	19.5	4.5	
Didelphys virginianus	.5	45.8	47.7	6.5	
Lynx rufus	.27	34.6	36.2	8.2	21.0
Canis latrans	.16	51.5	29.3	7.5	11.7
Capra aegagrus	.15	50.0	43.0	7.0	
Bos taurus	.10	50.0	44.0	6.0	
*Equus caballus	.10	74.8	19.9	5.3	

TABLE 3

RELATIVE GLAND SIZES, BY FOOD HABITS

Lynx rufus	.27	34.6	36.2	8.2	21.0
Canis latrans	.16	51.5	29.3	7.5	11.7
Didelphys virginianus	.5	45.8	47.7	6.5	
Capra aegagrus	.15	50.0	43.0	7.0	
Bos taurus	.10	50.0	44.0	6.0	
*Equus caballus	.10	74.8	19.9	5.3	
Dipodomys agilis	.67	76.0	19.5	4.5	

*Data obtained from Sisson and Grossman (1938)

PLATE I

Fig. 1. Goat Parotid: Schiff stain showing reactive material only in the connective tissues. Notice the large irregular lumen within acini. The parotid gland of the cow stains similarly. X 100

Fig. 2. Opossum Parotid: Schiff stain with negative reaction within parenchymal cells. X 100

Fig. 3. Kangaroo Rat Parotid: A mild Schiff reaction is produced, which is formed by fine granules within parenchymal cells. In the photograph the reaction can be seen as mottled gray areas. This was not seen in the parotid of another rat. X 100

Fig. 4. Coyote Parotid: A very strong Schiff reaction is present in the acinar cells. However, the photograph shows the reaction to be more intense than actual. X 100

Fig. 5. Bobcat Parotid: The intensity of the Schiff stain is similar to that of the coyote. Note the large amounts of connective tissue in the parotids of the bobcat and coyote. X 100

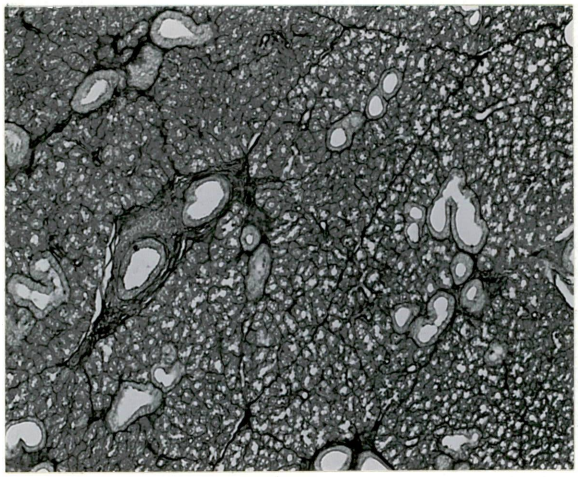

Fig. 1

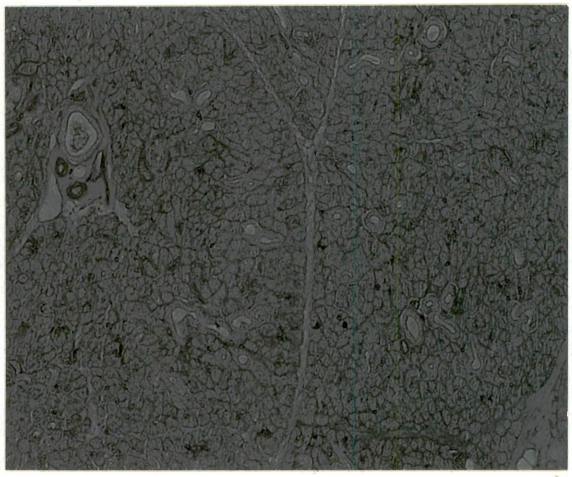

Fig. 2

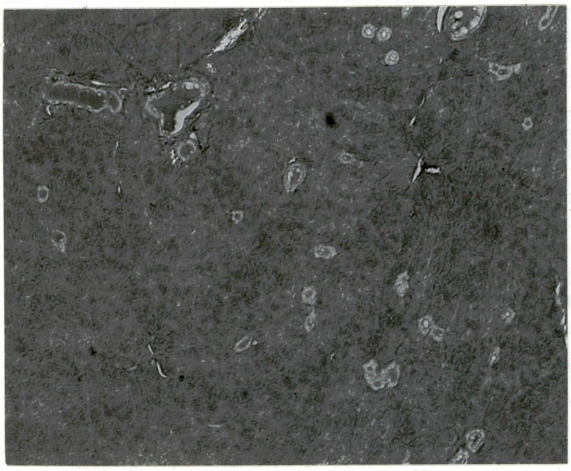

Fig. 3

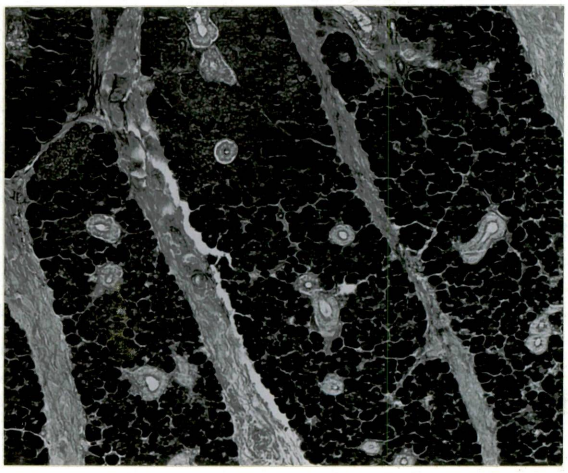

Fig. 4

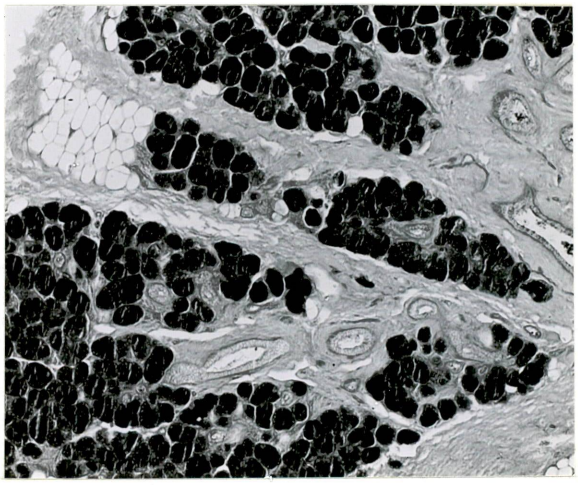

Fig. 5

PLATE II

Fig. 6. Coyote Submandibular Gland: An intense Schiff stain has darkened the mucous type cells. The demilune cells, which cannot be seen in this picture, did not stain. X 100

Fig. 7. Bobcat Submandibular Gland: Both demilune or serous type cells as well as mucous type cells have stained with the Schiff reagent. X 100

Fig. 8. Cow Submandibular Gland: The differences in precipitation of proteins by fixation produced the appearance of varied intensities of stain reaction with the Schiff reagent. The product of the mucous type cells was often reticulated, containing many clear spaces which caused the product to seem lighter staining than the serous type cells which had a homogenous texture. X 100

Fig. 9. Opossum Submandibular Gland: The unique system of ducts is seen clearly outlined by the dark areas of the Schiff stain. The pattern is different from that of any other gland because of spaces filled with large serous type cells which did not darken with Schiff reagent. X 100

Fig. 10. Goat Submandibular Gland: Two areas of different staining intensity are seen separated by a septum. The darker portion has few demilunes. It is largely composed of mucous acini. The activity of the ligher portion appears to vary. X 100

Fig. 11. Kangaroo Rat Submandibular-Sublingual Gland: The ligher portion reveals the duct system of the submandibular gland filled with P.A.S. positive granules. The mottling of the parenchymal cells is due to P.A.S. positive granules in the apex of the cells. The darker area shows the intense reaction of mucous acini of the sublingual gland. X 100

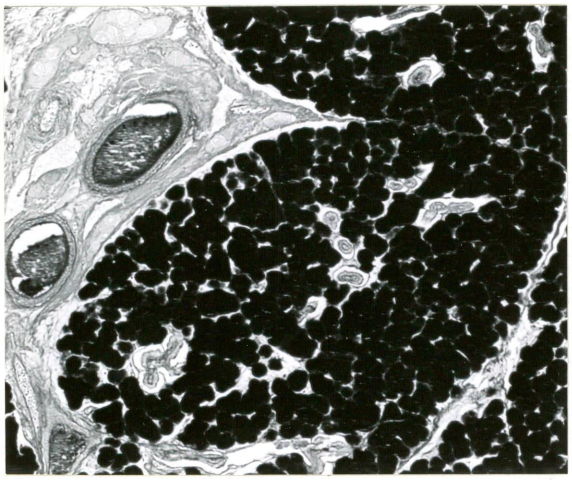

Fig. 6

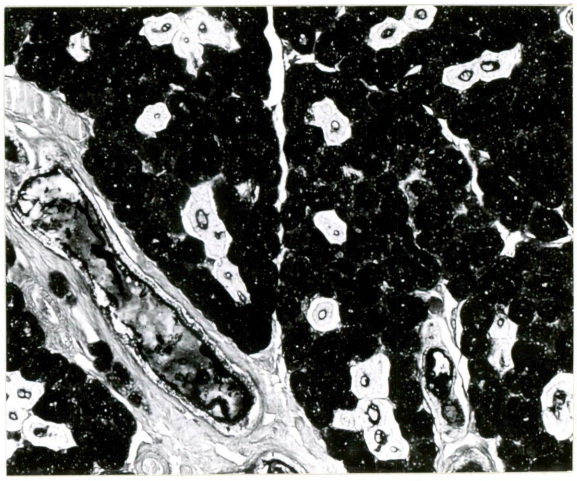

Fig. 7

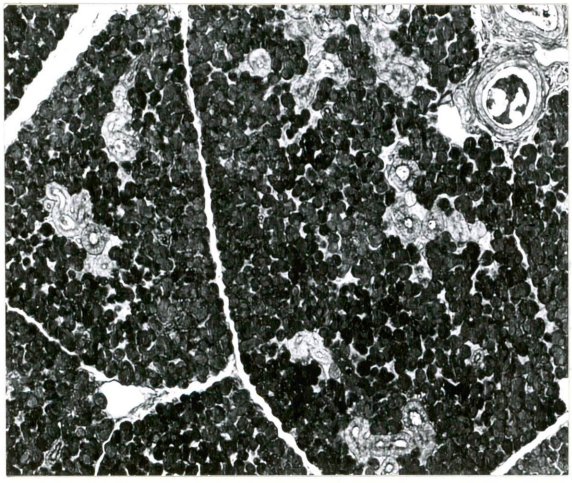

Fig. 8

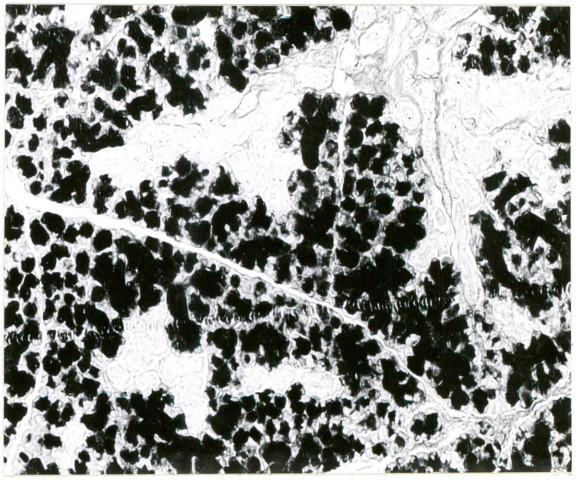

Fig. 9

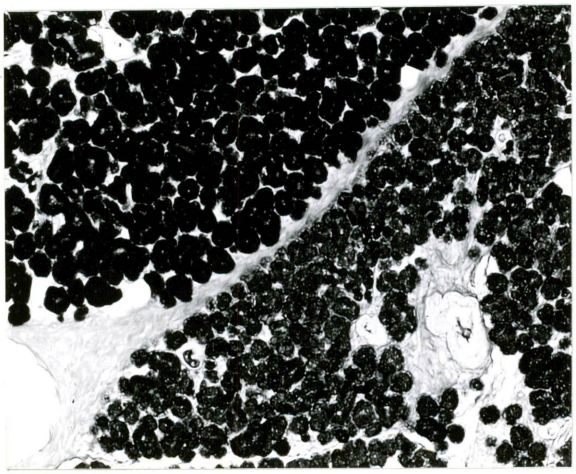

Fig. 10

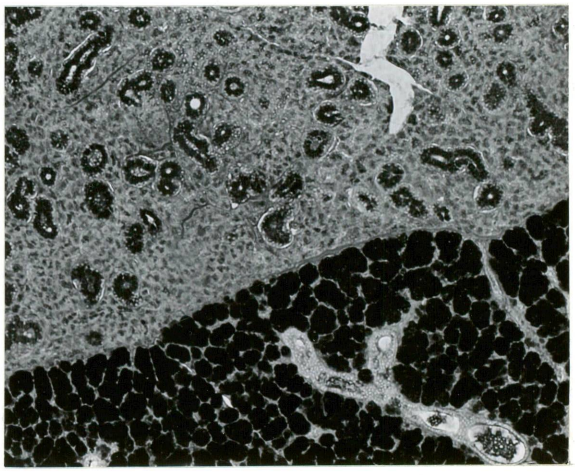

Fig. 11

PLATE III

Fig. 12. Coyote Sublingual Gland: The regions of intense staining are mainly the apical portions of what appears to be dark staining serous cells. The sublingual gland of the bobcat is similar. X 100

Fig. 13. Bobcat Infraorbital Gland: A view of the gland after Schiff stain. Note the long corridor lined by mucous type cells. X 100

Fig. 14. Opossum Sublingual Gland: The P.A.S. technique reveals a reactive substance in the apices of the cells. The basal parts of the cell do not stain. Note the large lumens in the tubular glands. X 100

Fig. 15. Cow Submandibular Gland: In the acini variations of intensity can be noted which may indicate differences in concentration or composition of the accumulated secretion product. X 100

Fig. 16. Bobcat Infraorbital Gland: Hematoxylin and triosin stain. Note the long corridors lined by mucous cells which empties directly into an interlobular duct. ID = interlobular duct. C = corridor. Approx. X 450

Fig. 17. Coyote Infraorbital Gland: Hematoxylin and triosin stain. The corridors are more branched in the coyote. In the photograph, the end of one indicated by arrow can be seen emptying into an intralobular duct. Approx. X 450

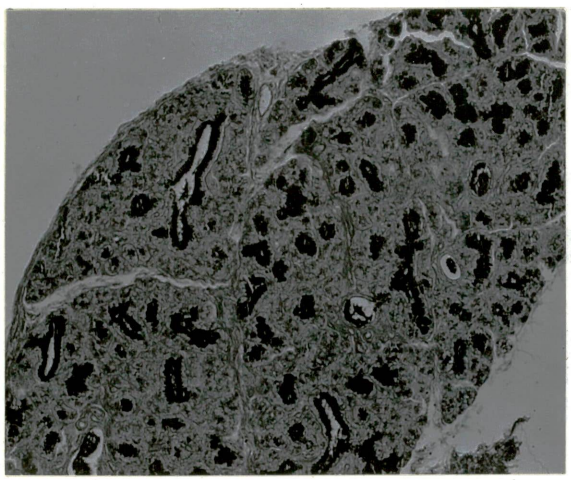

Fig. 12

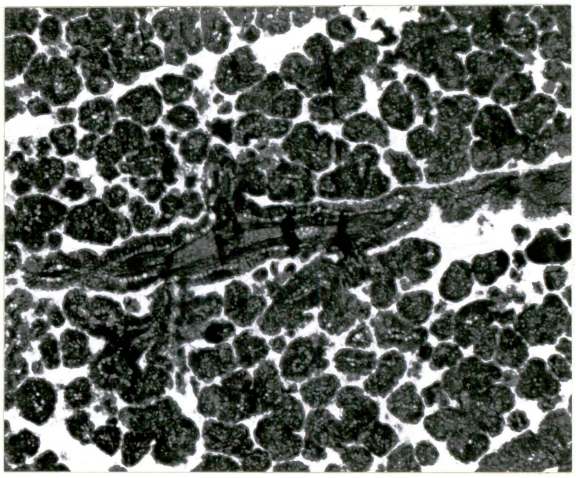

Fig. 13

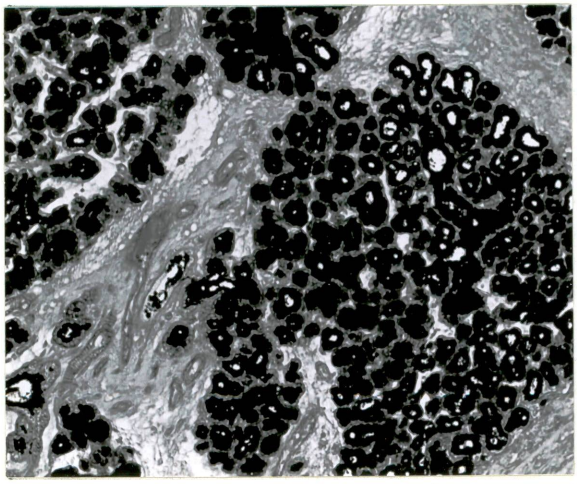

Fig. 14

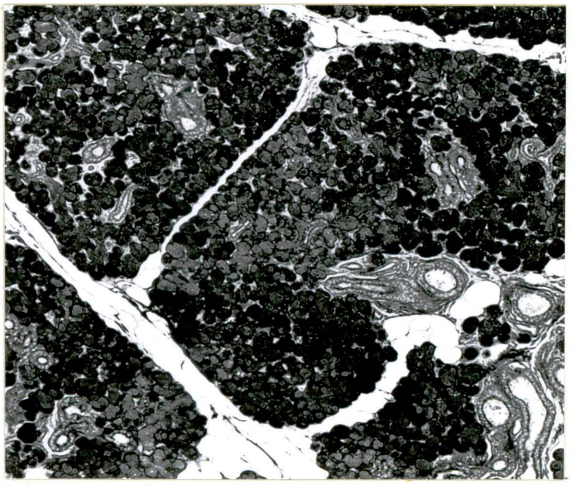

Fig. 15

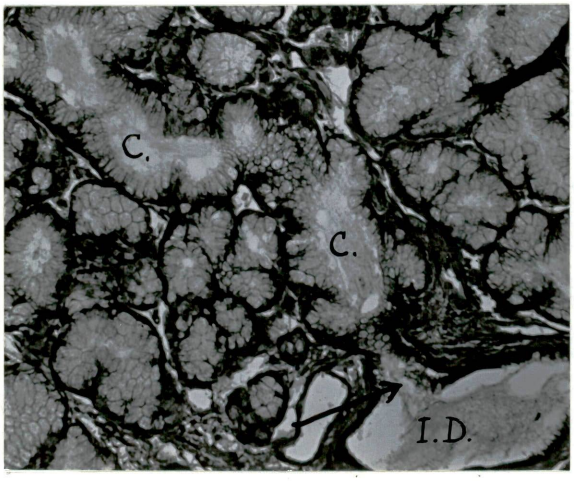

Fig. 16

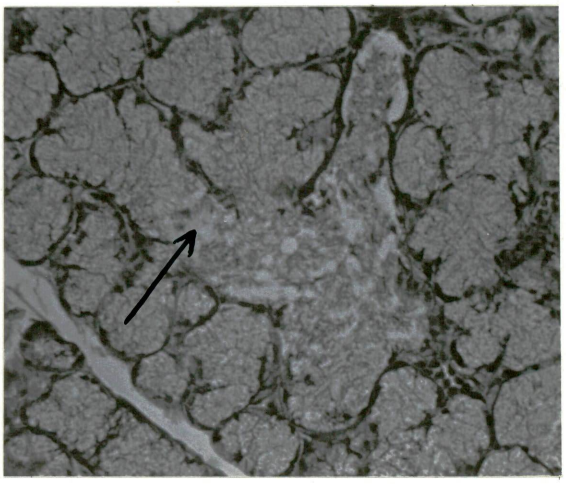

Fig. 17

PLATE IV

Fig. 18. Goat Sublingual Gland: Striking differences of intensity exists among cells within the same acinus. The variety of reactions suggest differences in composition or maturation of the secretory product. Approx. X 250

Fig. 19. Cow Sublingual Gland: An area of apparently reduced activity. Areas were noted having greater activity more nearly resembling the sublingual gland of the goat. Approx. X 250

Fig. 20. Cow Submandibular Gland: Highly magnified areas of cells in transitional stages. Arrows No. 1 indicate cells containing granules similar to those in the dark cells shown by arrow No. 2. The detail of the photograph is not sufficiently critical to fully convey the evidence. X 950

Fig. 21. Cow Submandibular Gland: The arrows point to cells in which the granules have nearly disappeared but can still be seen. In the lower right corner are fully distended mucous cells which do not reveal any granules. X 950

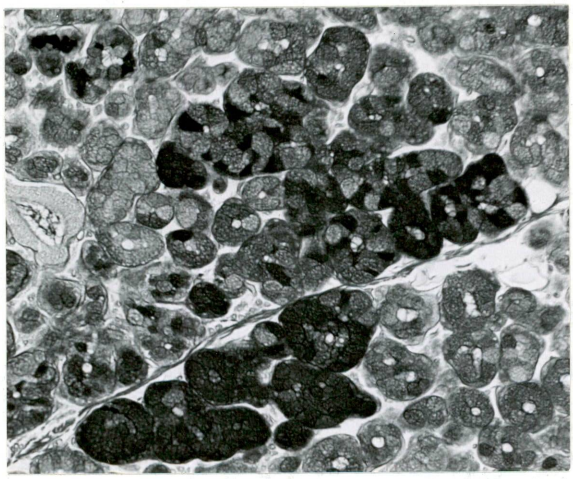

Fig. 18

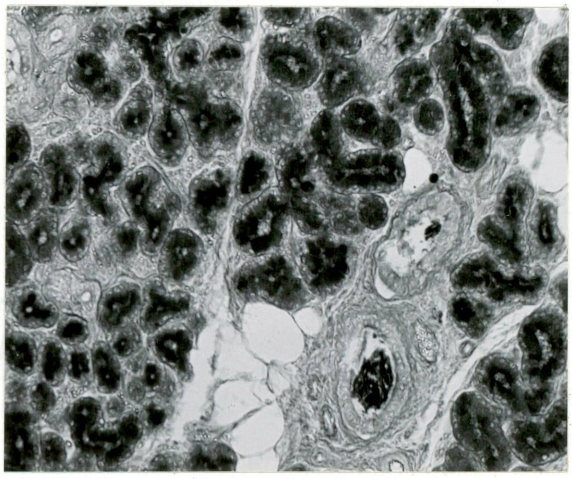

Fig. 19

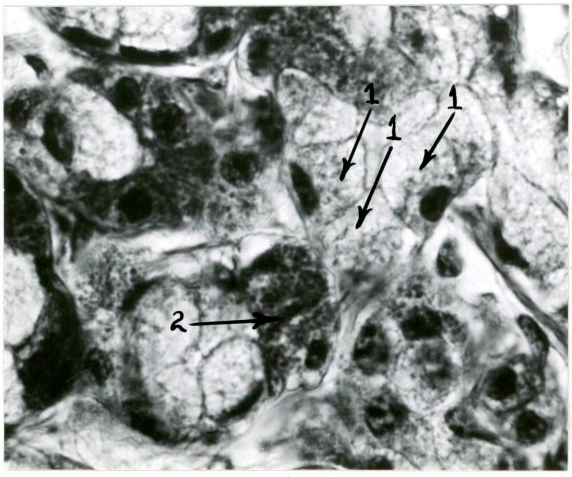

Fig. 20

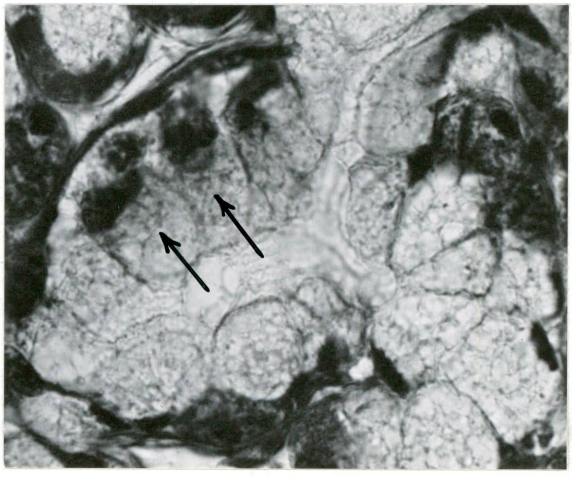

Fig. 21

PLATE V

Fig. 22. Opossum Submandibular Gland: Hematoxylin and triosin stain. A view of the compact arrangement of the duct system in the gland. X 100

Fig. 23. Coyote Submandibular Gland: Krischesky's modification trichrome. The dark demilunes are seen capping over the fine central lumen of the acinus. They are always found in such a position. Approx. X 950

Fig. 24. Kangaroo Rat Submandibular Gland: Hematoxylin and triosin stain. The arrow points to the nozzle-like arrangement of flattened cells penetrating the blind ending of a large serous tubule. The intercalated duct can be seen extending from the entrance for a distance. Approx. X 950

Fig. 25. Kangaroo Rat Submandibular Gland: Hematoxylin and triosin stain. A group of large serous tubules showing tall columnar cells and basally placed nuclei. Schiff reagent reveals the cytoplasm full of P.A.S. positive granules. Approx. X 950

Fig. 26. A diagramatic view of the intercalated duct system of the kangaroo rat as seen microscopically in fig. 24. The components are illustrated by: A, two zone staining acinar cells; B, lumen of branching intercalated duct surrounded by cuboidal cells; C, flattened epithelial cells which project into the lumen of the serous tubules; D, tall columnar epithelium lining the tubule.

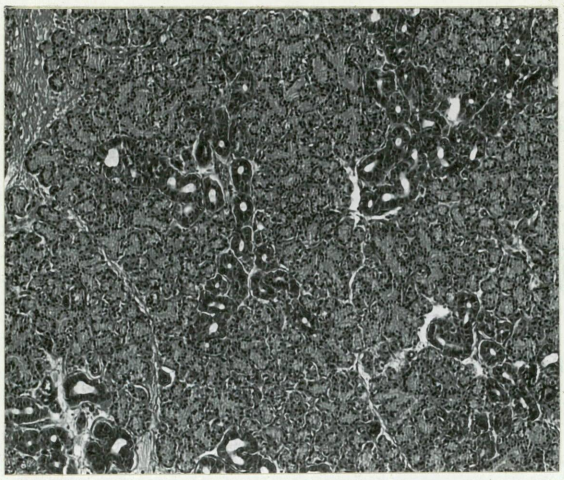

Fig. 22

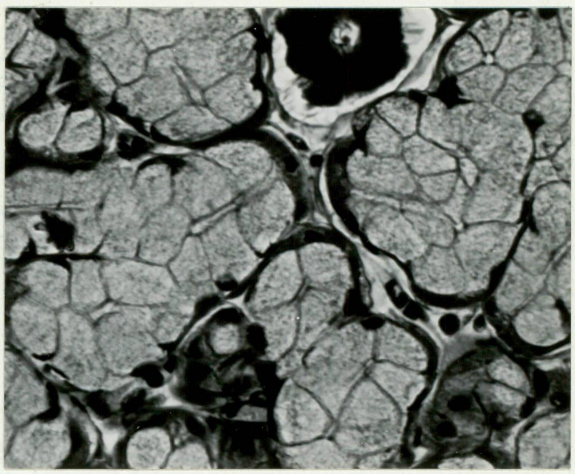

Fig. 23

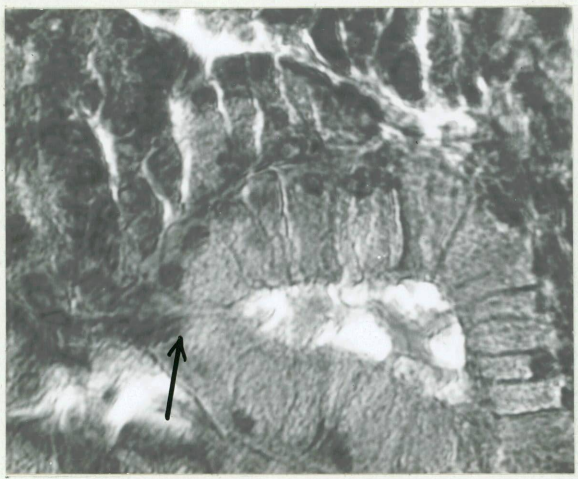

Fig. 24

Fig. 25

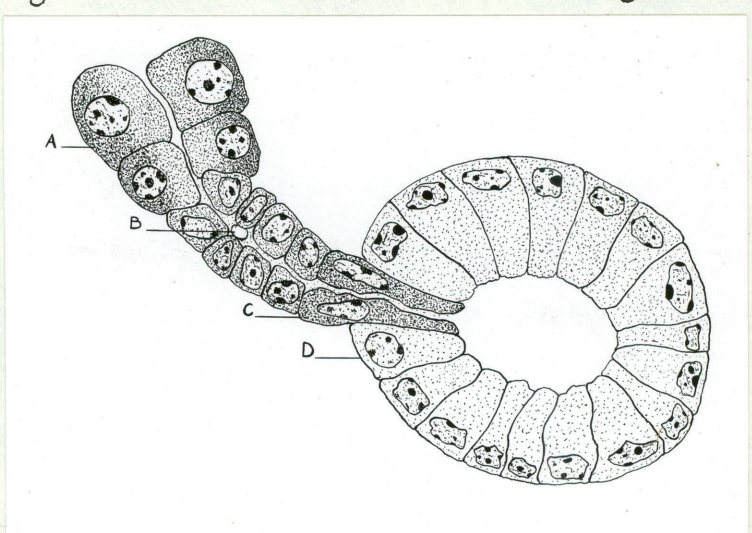

Fig. 26

PLATE VI

Fig. 27. Bobcat Submandibular Gland: A group of dark cells in a state of transition. It is suggested that they are a sero-mucoid type. Approx. X 950

Fig. 28. Bobcat Submandibular Gland: The arrow points to a demilune showing the general arrangement of the cells as a part of the wall of the acinus, each cell having an apex bordering the lumen. The light areas of the apices react positively with mucin stains. Approx. X 950

Fig. 29. Bobcat Submandibular Gland: An intercellular canaliculus can be seen at the end of the pointer between two cells. Each canaliculus is surrounded by a light zone devoid of the dark granules. Approx. X 950

Fig. 30. Opossum Submandibular Gland: The arrows indicate the arrangement of double membranes which surround all the cells of the acinus. Approx. X 950

Figs. 31, 32. Microscopic views of the parotid glands from two kangaroo rats of the same species. The cells in fig. 31 appear to be small, basophilic and empty of secretion. Those in fig. 32 are distended, lightly stained (more acidophilic) with nuclei pushed toward the bases of the cells. Approx. X 950

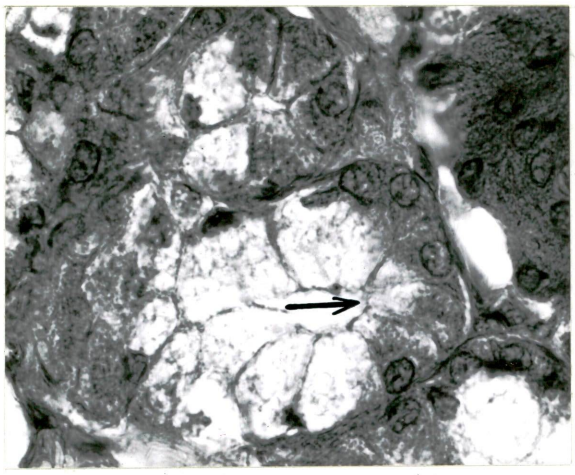

Fig. 27

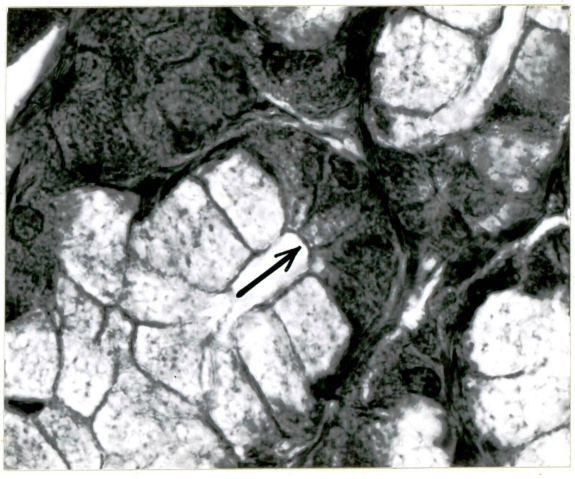

Fig. 28

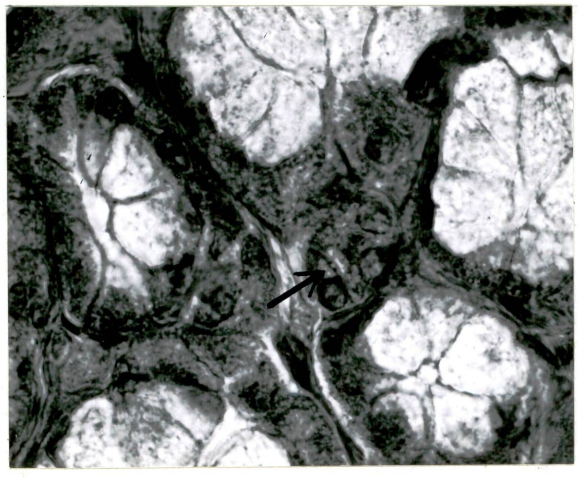

Fig. 29

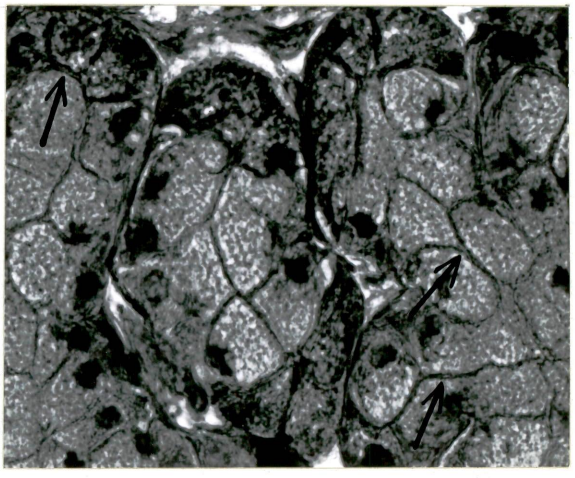

Fig. 30

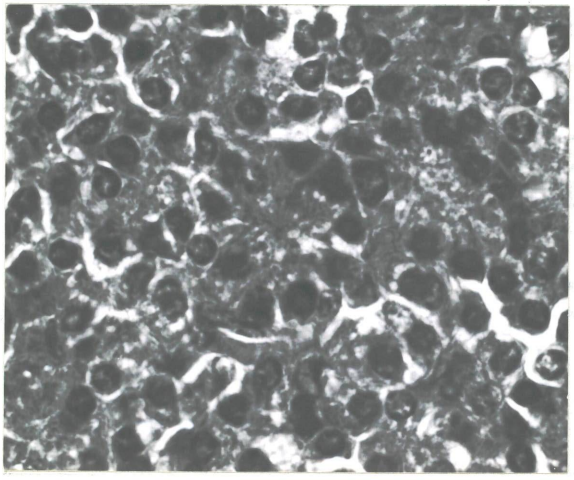

Fig. 31

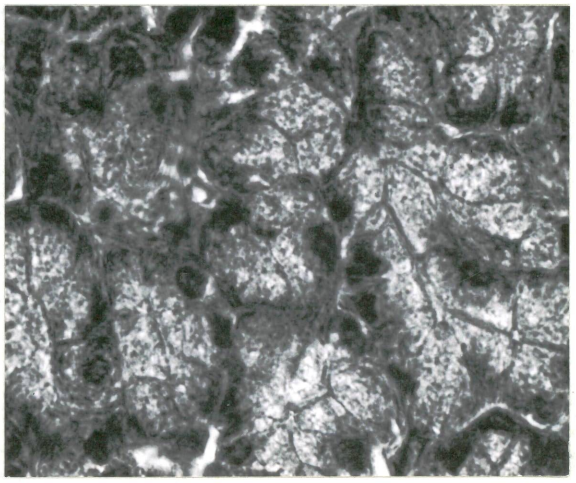

Fig. 32

COLLEGE OF MEDICAL EVANGELISTS

School of Graduate Studies

A COMPARATIVE STUDY OF THE SALIVARY GLANDS OF CERTAIN ANIMALS WITH WIDELY DIVERSE FOOD HABITS

by

Arthur E. Dalgleish

An Abstract of a Dissertation

in Partial Fulfillment of the Requirements

for the Degree Master of Science

in the Field of Anatomy

August, 1959

ABSTRACT

A comparative study was made of the salivary glands of six animals with diverse diet and food habits. These consisted of the following species: coyote, bobcat, opossum, kangaroo rat, cow and goat. Correlations were sought between the food habits of the animals and the relative size and microscopic structure of their glands.

After recording the gross weights of the animal and the removed glands, microscopic sections were prepared by the paraffin method. Tissues were stained by routine and special stains, including stains specific for mucins.

The cytology of the different glands and the varied mucin reactions are described. Special attention is given to certain anatomical features noted in this study which are not clearly described in the literature, such as the presence of intercellular canaliculi which may be generally present between mucous cells; and the unique arrangement of the intercalated duct system of the submandibular gland of the kangaroo rat.

The proportion of body weight made up by the salivary glands varies in these species inversely with total body weight. This may be related principally to surface-volume ratios and variations of metabolic rate rather than the type of diet.

The weight relationships of the individual glands are correlated more closely with the manner of eating than with the type of diet, particularly in the purely carnivorous or purely herbivorous animals. Failure of the weight ratios of different groups to conform to a linear distribution suggests other factors such as modifications of physiological functions and differences in secretory rates.

Study of the microscopic structure of the glands seems to reveal greater similarity among animals with close phylogenetic relationships than among animals with similar diets. The possibility that all secretory cells within a gland are of a single basic type is suggested and discussed.