THE CARDIAC GLANDS OF MAMMALS.

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WITH 16 TEXT FIGURES.

When we consider the immense amount of energy that has been expended on the investigation of the gastric glands since the discovery of the chief cells by Heidenhain and Rollet in 1870, it is a matter for some surprise that so little interest should have been aroused in the small zone of glands surrounding the cardiac orifice of the stomach, now generally known as the cardiac glands. This fact is all the more surprising when we remember that, in some mammals, these glands are by no means restricted to the narrow zone mentioned above, but may equal or even exceed in number the ordinary fundus glands composed of chief and parietal cells.

In examining the literature of this branch of research, it is difficult to decide to whom belongs the honor of discovering the glands in question, because the older anatomists employed the term to designate all the glands of the proximal portion of the stomach including the fundus glands. This example is followed in some recent text-books of Histology, the authors not recognizing that there are glands of more than two kinds in the stomach. In the interest of uniformity it seems desirable to employ the nomenclature adopted by Oppel, 96, in his compilation of the literature dealing with the stomach, and to designate the complex glands, composed of chief and parietal cells, fundus glands, reserving the expressions cardiac and pyloric glands for those which surround the respective orifices of the stomach.

Cobelli, 66, seems to have clearly recognized the cardiac glands in man as early as 1866, when he published a description in which they are referred to the same category as the pyloric glands, which he had studied some two years before, as gastric mucous glands.

The next contribution of importance is the account of the structure of the stomachs of two kangaroos, *Macropus* and *Dorcopsis*, published by Schäfer and Williams, 76, in 1876. These authors describe a very extensive region of the mucous membrane in these animals as being formed of simple glands, containing one kind of cell which resembles closely the cells of the submaxillary gland.

Ellenberger, 88, was the first to recognize the importance of the cardiac glands as indicated by their great extent in the pig, and the first to dissent from Cobelli's view that they are mucous glands. In his text-book he expresses the opinion that they are serous glands of a special kind, differing from both the fundus and the pyloric glands. This view is shared by Edelmänn, 89, the author of the most important contribution on the cardiac glands. Because of the importance of this work as containing the only comprehensive discussion of the cardiac glands, and because we owe to Edelmänn most of our knowledge of the nature and distribution of these structures in mammals, it seems desirable to quote at some length his conclusions:

"Im Magen der Säugethiere giebt es eine besondere Schleimhautregion mit belagzellfreien Drüsen, welche sich durch ihre Lage und durch histologische Eigenthümlichkeiten von der Pylorusdrüsenregion unterscheidet.

"Die Cardiadrüsenregion liegt entweder dort, wo die Schlundschleimhaut mit der Verdauungsschleimhaut des Magens Zusammenstösst, oder sie kleidet besondere Säcke aus oder liegt auch zum Theil isoliert in Vormägen ganz von cutaner Schleimhaut umgeben. . . .

"Eine Cardiadrüsenregion scheint bei den meisten Säugethieren vorzukommen. Sie fehlt sicher bei den fleischfressenden Cetaceen und den Wiederkäuern.

"Die Drüsen der Cardiadrüsenregion unterscheiden sich von der Fundusdrüsen nicht nur durch das Fehlen der Belagzellen, sondern auch von diesen und den Pylorusdrüsen durch die Anordnung und den Verlauf der Tubuli und die Eigenthümlichkeiten ihrer Drüsenepithelien."

This difference on which Edelmänn lays much stress consists in the fact that the cardiac glands subdivide at the neck of the gland, are arranged in groups, and have not so tortuous a course as the pyloric glands.

Edelmänn's conclusion that the glands are present in all Mammalia with the exception of the carnivorous Cetacea and the Ruminants, is opposed by Fleischmann, 91, who refers to their absence in three of the suborders of Rodentia, the Lagomorpha, Hystricomorpha and the Sciuromorpha, and concludes that they were certainly absent in the ancestral Rodents.

Further research, however, tends to support Edelmänn's conclusions as to their wide distribution in mammals and as to the great extent of the mucous membrane which they may occupy. For example, the investigations of Boas, 90, Pilliet, 85, and Cordier, 93, on the structure of the camel's stomach seem to indicate that the greater part of the cylindrical segment of the stomach in this animal is occupied by cardiac glands, and that the glands of the so-called water cells may properly be

referred to the same category. Cardiac glands have also been demonstrated by Oppel, 96, in *Dasyurus* and *Perameles*. The glands described by Salomon in the cœcal sac of *Cercopithecus* and *Inuus* are also in all probability cardiac glands, although the author did not recognize them as such.

Ellenberger, 88, not only dissented from the commonly accepted view that the cardiac glands are mucous glands, but went farther and attempted a solution of the question of their physiological rôle. He found, in conjunction with Hofmeister, 85, that the cardiac glands of the pig contained a diastatic ferment. Negrini, 86, also asserts the presence of an amylolytic ferment in the cardiac mucosa of the pig, in greater quantities than could be accounted for by the amount of blood present in the organ. The results of Ellenberger and Hofmeister are confirmed by Edelmann who tried many of the so-called mucin stains with negative results. Edelmann's explanation of the function of the cardiac glands is as follows:

"Die physiologische Bedeutung der Cardiadrüsenregion beruht in der Bildung einer Art Vorraum im Magen, welcher keine Säure, dagegen Fermentquellen enthält, und in dem die Verdauung der Stärke vor sich gehen kann." "Als Schleimbildende Drüsen sind die Cardiadrüsen nicht aufzufassen."

Schaffer, 97, also, in his recent studies on the human cardiac glands, ranges himself on the side of Edelmann and Ellenberger as to the serous nature of the cardiac glands. Using the more precise mucus stains recently devised by Mayer, he was unable to obtain any reaction in the cells of the cardiac glands, which he, therefore, compares to the chief cells of the fundus glands, and to the pyloric gland cells.

There is some danger of attaching too much importance to these negative results of Schaffer and Edelmann. In the first place, mucin is not a single chemical substance for which it is possible to establish definite staining reactions, which will enable us to demonstrate it wherever it occurs. There are on the contrary good reasons for supposing that, as Huppert, 96, points out, the number of mucins belonging to a single class, the glycoproteids, are as numerous as the albumens which are available for entering into such a combination with a sugar. In the second place, the morphological evidence adduced by Krause, 95, and Langley, 84, in the salivary glands and the chemical experiments of Hammarsten, all point to the conclusion that there are several stages in the elaboration of mucins in secreting cells. It is probable also that there exist in mucin-forming cells differences of secretory equilibrium analogous to those which have been noted by Langley in pepsin-form-

ing cells, and that, for example, one cell may store its mucin in a more elaborated form than another cell which is its morphological equivalent. It is unreasonable to expect that the different mucins and different stage of elaboration of mucin would present always the same staining properties.

The importance of this point of view is clearly recognized by Mayer, 97, in his recent paper "Ueber Schleimfärbung" in which he points out that, in relation to his muchaematein solution, it is possible to construct a graded series beginning with mucins which stain only with difficulty and passing by imperceptible gradations to mucins which stain rapidly and deeply. He was also able to obtain in the cells of the submaxillary gland of the hedgehog, the secretion of which, according to Krause, does not contain mucin, a typical mucin reaction.

It may be pointed out, also, that the obtaining of mucin reactions in secretory cells is to a certain extent dependent on the technique of the individual investigator. The writer, for example, as will be indicated in the more special portion of this paper, has had little difficulty in staining the cells of the cardiac glands in every instance with mucicarmine and muchaematein, and, in some cases, with other mucin stains such as indulin and methyl blue.

It is clear from the foregoing that *negative* results with mucin stains are absolutely without value as evidence of the serous nature of secreting cells, and that positive results are only of value in so far as they are confirmatory to evidence derived from other sources. Some caution is even necessary in drawing conclusions from the structure of the cell. The submaxillary gland of *Erinacæus* is a case in point, where the cells have both the structure and staining properties of mucous cells, although no mucin is secreted by the gland. Another case is that of the pyloric gland which was for many years interpreted as a serous gland, simply because the nucleus is often spherical instead of crescentic, and because the cells may contain considerable residual protoplasm.

Edelmann's theory of the function of the cardiac glands would be a simple and sufficient explanation of the occurrence of these glands in herbivorous mammals, but for the extraordinary fact that they are absent or feebly developed in the most highly specialized of herbivores, the ruminants. The occurrence of large areas of cardiac glands in all Perissodactyls and in the pigs, camels and llamas among the Artiodactyls, precludes the possibility of supposing that they were absent from the stomachs of the herbivorous ancestral types, in which their original occurrence is rendered more probable by the discovery by Zimmermann and Sal, 94, of what may prove to be remnants of cardiac glands along

the œsophageal groove in the sheep. If the glands are really of importance as sources of a digestive ferment, it is difficult to understand why they have disappeared in a group of animals in which their presence would be such an immense advantage. One would rather expect that the processes of natural selection would tend to preserve and perfect them. The importance of the occurrence of an amylolytic ferment in the cardiac mucosa is materially reduced by many recent researches which go to show the widespread occurrence of sugar-splitting ferments in the blood and tissues of mammals, and by the observation of Ellenberger and Hofmeister that a diastatic ferment is present in the fundus mucosa of the pig where, owing to the acidity of the gastric secretion, it could be of very little service in digestion.

In the present state of our knowledge it does not seem possible to assign any definite function to the cardiac glands, nor does investigation by physiological methods appear to offer much hope of solving this problem. It seems to the writer that the first step in the further elucidation of these glands must consist in the comparison of them, as regards structure, staining, and microchemical reactions and mode of regeneration, with the other glandular elements of the stomach. It is true that both Edelman and Schaffer have instituted comparisons of this kind, but at the time of the publication of Edelman's paper, no adequate description of the structure of the chief cells and pyloric cells existed, and Schaffer's comparison of the cardiac gland cells with the chief cells as well as the pyloric gland cells shows clearly that he was unaware of the important differences between the two.

Another problem which must be considered in connection with the question of the histology of the glands is that of their phylogeny. The fact that only fundus and pyloric glands are represented in reptiles and batrachians makes the question of the source of the cardiac glands a very interesting one. Are they to be regarded as derived from pre-existent structures, for example, as œsophageal glands which have been taken over into the stomach, or fundus glands which have been peculiarly modified, or finally as cœnogenetic structures which have arisen in Mammalia in response to a new functional demand? Oppel, 96, has suggested, but in a tentative rather than an assertive mood, that they may represent the simple glands of lower vertebrates, which have not all been differentiated into the complex fundus glands of mammals, but admits that the great structural differences are scarcely compatible with such a view. Edelman contents himself with stating their possible sources of origin without attempting to decide between them. More recently Oppel, in a brief discussion of Schaffer's discovery of parietal

cells in the cardiac glands of man, suggests the possibility that these structures may have been derived from the fundus glands by the gradual disappearance of the parietal cells and that this change may be the initial phase of the process by which the glands are replaced by stratified epithelium.

It will be observed that, with the exception just noted, the only attempts which have been made to explain the occurrence of cardiac glands in mammals have been based on the *a priori* assumption that they have a useful function to perform in digestion. The alternative hypothesis that they constitute a stage in an advancing process, and that their production and subsequent disappearance in some forms is the result of the continued and consistent action of the same causes, has scarcely been discussed at all.

The basis for comparison adopted in this paper is the structure of the chief cells as described by the writer and confirmed by Zimmermann, 98, Cade, 01, and others. In 1896, in a preliminary note, 96, and subsequently in a more extended paper, 98, I pointed out that the chief cells of the body of the fundus glands in common with certain other serous glands possess structural and microchemical characters which enable one to distinguish with ease between them and all other glandular elements of the stomach. These are briefly as follows: The cell is divisible into a proximal and distal zone. The distal zone next the lumen contains granules of zymogen inclosed in protoplasmic trabeculae. The proximal zone, of variable extent at the attached end of the cell, exhibits an indistinct radial striation (basal filaments of Solger), and stains strongly in nuclear dyes, owing to the presence in it of a kind of chromatin, prozymogen, which, like the chromatin of the nucleus can be shown by Macallum's microchemical methods to contain iron in a masked form as an organic compound. MacCallum, 98, has since shown that this substance as well as the zymogen granules contains phosphorus. I found that the chief cells of the neck of the gland were the homologues of the large clear cells of the corresponding portion of the fundus gland of lower vertebrates and of the cells of the pyloric glands, all of which differ in fundamental points from the ferment-forming chief cells of the body of the gland. They contain neither basal filaments nor granules of zymogen and their protoplasm gives only a faint microchemical reaction for iron. These neck chief cells and pyloric gland cells I found to pass into the surface epithelium by a gradual transition, the mean of which is reached at the deep end of the gastric pits or foveolae where actively dividing cells are found which are probably engaged in replacing, in accordance with the "wander"

theory of Bizzozero, the surface epithelium and to some extent these peculiar neck cells and pyloric gland cells. Because of this relationship to the surface epithelium, which is undoubtedly muciparous, and because of the fact that it stains readily with Mayer's mucin stains and with certain other distinctive stains such as indulin, I regard the secretion of the neck chief cells and pyloric gland cells as of a mucous nature.

The first section of this paper will be devoted to the extension of the same methods of research to the cardiac glands. The second section will be devoted to a discussion of the phylogeny of the cardiac glands and of some points in the evolution of the complex forms of mammalian stomachs, with which, in the writer's opinion, these glands are intimately connected.

THE HISTOLOGY OF THE CARDIAC GLANDS.

I. THE CARDIAC GLANDS OF MAN.

I have chosen the cardiac glands of man for preliminary description because of the importance of the structure of the human stomach from the physiological and pathological standpoint and because of the fact that an opportunity, like that which I have fortunately met, so rarely presents itself of securing the normal gastric mucous membrane of man in a good state of preservation. The cardiac glands of man also illustrate very well the points which I regard as of histological and phylogenetic importance. My material was secured at a necropsy on the body of an executed criminal, a young man of about thirty years of age, at which I was enabled to be present through the kindness of Professor A. Primrose. Pieces of the mucous membrane of the cardiac orifice and from the important regions of the stomach were fixed in alcoholic bichromate sublimate and in absolute alcohol. The whole of the regions surrounding the cardiac and pyloric orifices were similarly fixed in the bichromate sublimate mixture and the rest of the stomach then preserved in seventy per cent alcohol for topographical study. On examination, all of this material proved to be in an excellent state of preservation although the superficial epithelium was lost in some places and somewhat altered in others. The necropsy occurred about forty-five minutes after death.

The cardiac glands of man have been recently the subject of a very careful research by Schaffer, 98, whose descriptions, in so far as they are concerned with the topography and general structure of the glands leave little to be desired. According to Schaffer the cardiac gland zone begins from one-half to four mm. above the termination of the œsophageal epithelium, and extends a distance into the stomach which

varies with different individuals and in different portions of the zone in the same individual. The maximum extent is given by Kupffer, 83, as 1.5 cm., the minimum as .5 cm. That the maximum may be much greater than this is indicated by the present case in which the cardiac glands begin nearly 3 mm. above the termination of the œsophageal epithelium and extend a distance of 4.3 cm. into the stomach. The latter measurement is following the various folds of the mucous membrane, the length in a straight line being 3.4 cm.

The mucous membrane at different points in this region is of very unequal thicknesses, the differences in this respect being partly due to the size and mode of aggregation of the glands, partly to the number and size of the lymphatic follicles which, as Schaffer indicates, are very numerous. In general it may be said that the greatest thickness occurs at or near the œsophageal epithelium and the minimal at the point where the cardiac gland zone passes into the fundus gland zone. In a zone comprised within a distance of 2 mm. from the œsophageal epithelium the thickness of the mucous membrane, not including the muscularis mucosæ, varies from .99 mm. to 1.326 mm. Three millimeters from the œsophagus in one section which is fairly representative the thickness is .969 mm. From this point onward there is a gradual diminution in thickness until at a point 11 mm. from the œsophageal epithelium the minimum thickness of .5 to .6 mm. is reached, which is maintained, with slight variations probably due to different degrees of contraction of the intrinsic muscle fibers, for the rest of the cardiac zone.

This great difference in the relative thickness of the mucous membrane of the proximal and distal portions of the zone is due to a difference in the nature of the glandular aggregates, not to a fundamental difference in their structure. Just at the termination of the œsophagus, the glands consist for the most part of freely branched and tortuous tubules, derived from a narrow duct which ascends in the direction of the free surface of the mucosa to open alone, or more frequently in common with others, into a depression of the surface which corresponds to the ducts or foveolæ of the glands of the rest of the stomach. Several such tubulo-racemose glands are commonly grouped together into a sort of lobule more or less definitely separated from adjacent groups by a sort of septum of collagenic fibers containing bands of smooth muscle. The ducts of these glands are frequently the seat of retention cysts which may be of such a size as to be readily recognized without the aid of a microscope. These may occur near the opening of the gland into the foveolæ or deeper in the mucosa. The cysts appear as large oval ampullæ into which the branches of the gland open. The glands and ducts are lined throughout by secreting cells.

As the distance from the œsophageal epithelium increases and the mucous membrane diminishes in thickness the tubules become less and less branched and the peculiar grouping disappears, although an apparent grouping may sometimes be observed, caused by the regular occurrence at intervals of lymphoid follicles. The cysts also become rare and the number of foveolæ opening on a given area more numerous. In the distal thin portions of the mucous membrane there is little difference between the cardiac glands and the adjacent fundus glands as regards their shape and mode of branching.

FIG. 1. Longitudinal section of the tunica mucosa of the cardiac region of man, 2.5 cm. from the edge of the œsophageal epithelium. $\times 70$. The leucocytes and plasma cells are considerably more numerous than in the figure.

Throughout the area there is considerable interglandular tissue of a reticular character, the meshes of which are filled with leucocytes of various types. Considerable collagenic tissue is also present between the bases of the glands and the muscularis mucosæ. Many bands of smooth muscle fiber ascend from the region of the muscularis mucosæ in the direction of the epithelium. The structure of this interglandular tissue will be discussed at greater length in a paper on the "Structure of the Human Stomach," now in course of preparation.

Lymph follicles are very numerous in the proximal thicker portion. They occur at fairly regular intervals and are situated in the mucosa,

but as the membrane becomes thinner they become first restricted to the deeper portion of the mucosa and finally break through the muscularis mucosa to extend into the submucosa. In the latter case the muscularis mucosæ is interrupted opposite the follicle, the superficial tissues of which are continuous through the gap with the diffuse adenoid

tissue of the mucosa (Fig. 1).

There are reasons, which will be discussed later, for regarding the cardiac glands as decadent structures. On this account it is desirable to select for preliminary description those glands in which this feature is least apparent. Such are the glands of the thinner distal portion of the mucous membrane of the cardiac zone. To avoid confusion, I shall call the funnel-shaped depressions of the surface into which the glands open, foveolæ, as being a preferable term to the compound expressions gland-duct

FIG. 2. Two foveolæ gastricae from the cardiac region of man, $\times 280$. The mucous theca becomes reduced, in the lower half of the foveola, to a very narrow margin along the lumen. The cells in this position are shorter and a number of deeply staining mitotic figures are seen. A few polymorphonuclear leucocytes and lymphocytes are to be seen in the epithelium. The cells of the interstitial tissue are largely Unna's plasma cells with a few polymorphonuclears and eosinophiles.

and stomach-pit, commonly employed.

The epithelium of the free surface was lost in most of the material at my disposal, but where it was retained resembled in all essential points that of other stomachs. The epithelium of the upper portion of the foveola is represented in Fig. 2 and is very similar to the epithelium of

similar portions of the stomachs of other mammals. The cells are elongated conical structures. A well-defined mucous theca occupies the outer third of the cell, an oval nucleus the inner protoplasmic portion. Passing down the sides of the foveolæ there is observed a gradual reduction in size, from cell to cell, of the mucigenous border, and for a variable distance at their lower ends foveolæ are lined by columnar cells in which only a very narrow band of mucin can be distinguished along the free border. The spherical or oval nuclei of these cells are, as compared with the nuclei of the cells of the surface epithelium and those of the glands, exceedingly rich in chromatin and are frequently seen in the process of mitotic division, from one to four or even more mitoses being seen in each foveola. Many leucocytes, both lymphocytes and polymorphonuclear cells, could be seen in my material, within the epithelium of the foveolæ, in most cases occupying spaces between the cells but occasionally intracellular.

For a short distance at their upper ends, the glands are lined by cells similar in all respects to those of the deep ends of the foveolæ, but at a short distance from the foveola the cells begin to give evidence of greater activity in secretion inasmuch as the portion of the cell so engaged increases in size and the protoplasmic portion containing the nucleus becomes correspondingly reduced. This feature becomes more and more prominent as the bottom of the gland is approached, where the cells may be entirely filled with secretion. In such cells the nuclei are compressed, flattened or crescentic, and are found at the base of the cells. As a rule, there is considerable variability in the cardiac glands as to the degree of loading exhibited by the individual cells of a terminal tubule or of different terminal tubules, some cells being completely filled with secretion, others only partly so. In the latter type, the cell is sharply divided into two zones, a proximal protoplasmic zone in which a fine reticulum may be seen, and a distal zone which appears transparent in ordinary preparations and contains the stored-up secretion (Fig. 3, A).

The outer zone does not contain basal filaments, nor is it markedly chromophile. Examined for masked iron by the usual methods, a slight reaction is obtained, but no greater than that exhibited by the protoplasm of the cells of the surface epithelium, the pyloric gland cells, and the neck chief cells of the fundus glands. The inner zone of these cells stains but faintly in hæmatoxylin and eosin, but one can readily recognize a wide-meshed alveolar structure, the meshes of which correspond to the protoplasm between the droplets of secretion. The apparent thickness of these protoplasmic meshes is increased by the precipitation on them of the solids of the secretion. Frequently the

mass of secretion contained within a cell is incompletely divided into two masses by a strand of reticular protoplasm stretching across the cell (Fig. 3, A). In such cases the proximal mass of secretion usually contains a protoplasmic network composed of thicker trabeculæ than the distal mass. This transverse band is particularly obvious in specimens stained by Heidenhain's iron hæmatoxylin method and is of frequent occurrence in all the mucin-forming elements of the stomach as well as in the cells of the glands of Brunner (Zimmermann,

FIG. 3. Three tubules from cardiac glands of man. $\times 1000$. A, Mucous tubule in which cells are in early stage of mucin formation. B, Mucous tubule showing cells in various stages of mucin formation. Subdivision of mass of secretion into two masses by transverse bridge of protoplasm and the gradual disappearance of the latter as the proximal mass increases in size. C, Tubule formed of cells of the type of the chief cell of the body of the fundus gland, showing zymogen granules in the distal zone and prozymogen (basal filaments) in the proximal zone. D, Binucleate parietal cell with intracellular secretion channels.

98) and in mucous cells from other sources, Maximow, *op. cit.* As the cell fills with secretion (Fig. 3, B), the distinction above referred to between the protoplasmic framework of the two masses of secretion gradually disappears and the transverse band which separates them becomes disintegrated and may finally disappear, although, as a rule some slight re-

mains of it may usually be recognized in the interior of the cell. In some cases the proximal mass of secretion only is present so that the cell has a protoplasmic distal border along the lumen, and reminds one strongly of Krause's figures of the mucin-forming cell of the retro-lingual gland of *Erinaceus* at an early stage of mucin formation.

The mitoses seem to be entirely confined to the cells of the bottom of the foveolæ and adjacent portions of the gland. I have not observed a single instance of cell division in the surface epithelium nor in the cells at the bottom of the gland, although cells near the foveolæ may divide even when they contain a good deal of secretion. The great mitotic activity at the junction of gland and foveola, as well as the gradual transition from this point in both directions leads me to believe that this is the site of the reproduction of both the surface epithelium and the glandular epithelium, both of which are probably replaced when lost by a gradual migration of cells from the point at which they are reproduced.

Passing on, now, to the complex glands of the beginning of the area this simple relation of the cellular elements to one another is found to be by no means the rule. Many anomalies are seen. The cells of the lower ends of the foveolæ while similar in the majority of cases to those already described may lose their mucigenous border altogether and appear protoplasmic throughout. Again the transition into the gland may not occur at all or be little marked, the secretion filling only a small portion of the free border of the cell even in tubules remote from the main duct. Thus, one may see side by side in the same gland, cells or tubules equally remote from the duct which exhibit the two extremes of secretion, some filled with stored-up product, others containing but little. These are, obviously, differences of physiological equilibrium and do not introduce any new difficulties in the way of interpreting the glands. In this region the mitoses, as in the case of the more typical glands, are practically confined to the upper portions of the ducts and the adjacent portions of the gastric foveolæ. There is also in the complex glands near the œsophageal epithelium a great variability in the size of the terminal branches of the tubules which compose the gland, some being many times as large as others.

The important question now arises, what is the nature of the stored-up secretion in these cells? The transition to the surface epithelium and the probable mode of reproduction of the glandular elements as indicated by the mitoses, both suggest that the secretion may be chemically, as the cell is genetically, related to that of the surface epithelium, that is to say, may consist largely of mucus. On the contrary Schaffer,

97, and Edelmann, 89, agree that the contents of the cells do not stain with mucin stains. In view of the fact that Schaffer used for this purpose mucicarmine and muchæmatein as recommended by P. Mayer, and that my results with similar solutions are directly opposed to his, inasmuch as I succeeded in staining the secretion in the cells of the cardiac glands as well as those of the surface epithelium, it may be of interest to discuss briefly the reasons of the discrepancies in our results. Some reference has already been made to the probably great chemical diversity of the mucins and to the work of Mayer on their staining properties. In the course of this paper he says:

“Ich selber habe bisher das Wort Mucin geflissentlich vermeiden und dafür stets von Färbung des Schleimes gesprochen. In der That wissen wir vom Schleime der höheren Thiere chemisch noch recht wenig und von dem der allermeisten Wirbellosen so gut wie gar nichts. Oben habe ich bereits angedeutet dass die Schleime sich gegen einen und denselben Farbstoff sehr verschieden verhalten, indem ich bei Besprechung der Lösungen von Hämatoxylin oder Hæmatein sagte: In der Regel färbt sich der Schleim, oder: in der Regel färbt er sich nicht. Das sollte heissen: die von mir geprüften Arten Schleim sind je nach ihrer Provenienz verschieden. Man könnte da eine förmliche Reihe aufstellen, die mit solchem Schleime zu beginnen hätte, dessen Färbung kaum zu verhindern ist, und mit solchem enden würde, der sich kaum noch färben lässt. Jener ist z. B. bei Muscheln vertreten dieser findet sich im Darm von Homo.”

I have found that not only do different mucins stain with different degrees of readiness in Mayer's solutions, but that by modifying the composition of the solution or the mode of applying it, many mucins which stain with difficulty in the ordinary method may be made to take the color readily. For example, if a longitudinal section in celloidin of the pylorus of man be placed in Mayer's muchæmatein (Hæmatein .2 grammes, aluminium chloride, .1 gramme, 70% alcohol, 100 cc.), only the goblet cells of the surface epithelium and of the glandulæ intestinales stain. If the strength of the solution in hæmatein and aluminium chloride is doubled, then the goblet cells stain strongly and a faint reaction is obtained in the epithelial cells of the stomach and of the pyloric foveolæ. If the strength of the solution is increased fivefold, so that it contains one per cent of hæmatein and one-half per cent of aluminium chloride, both the secretion of the goblet cells in the intestine, and of the epithelium in the stomach, stains intensely and an equally vigorous stain is obtained in the cells of the duodenal glands (glands of Brunner) and in the cells of the pyloric glands. Practically the same result was obtained with mucicarmine. With the diluted solution recommended by Mayer, only goblet cells were stained, but if

the strength of the solution were increased, the gastric epithelium stained. With Mayer's undiluted stock solution of mucicarmine, we find in addition the pyloric glands and duodenal glands stained. Thin sections from which the imbedding mass has been removed and which are allowed to float freely in the solution will give positive results with much less concentrated solutions than sections in celloidin, or sections attached to the slide. For example, I obtained positive results in the pyloric gland cells of the cat and in the neck chief cells of the fundus glands of the same animal, by simply transferring thin sections, cut after imbedding in paraffin, from alcohol to the diluted solutions recommended by Mayer.

If the undiluted mucicarmine solution of Mayer or the muchæmatein solution of fivefold strength be employed, positive results may be obtained with perfect certainty in the glandular cells of the pyloric glands, the duodenal glands of Brunner, the cardiac glands, and in the neck chief cells of the fundus glands of a great many mammals. Material fixed in alcohol or in bichromate sublimate has yielded the best results in my hands, but other fixing agents do not materially alter the character of the reaction. Sections cut in celloidin stain perfectly in the above solutions, although the time of staining with muchæmatein must not be too prolonged or the celloidin mass will take the stain. With mucicarmine, the celloidin stains somewhat, but in the tissues the color is entirely confined to the secretory contents of the cells.

In cardiac glands so treated, the inner portion of the cell, which remains unstained in sections stained by other methods, is deeply colored blue or red according to whether muchæmatein or mucicarmine is employed. The protoplasm of the cells and the nuclei remain unstained. Schaffer's failure to obtain this reaction was probably simply due to the fact that his solutions were too dilute or that he did not take advantage of the greater rapidity of action of the dilute stains on sections unattached to the slide and free from any imbedding mass.

There can be very little doubt that the cardiac gland cells of man are muciparous structures. Their relation to the cells of the surface, the staining reactions which I have observed, the compression phenomena observed in the nuclei in the completely filled cell, and the absence from them of zymogen granules and prozymogen (with certain exceptions to which I shall refer presently) all clearly point to this. I should hesitate to assert, however, that this is the only kind of activity in which the cardiac gland cell is engaged although, indeed, it is the most conspicuous and impresses itself most clearly on the structure of the cell.

A point remains to be discussed which is of the greatest importance in deciding the origin of the cardiac glands, namely, the occurrence in them of the other characteristic elements of the fundus gland, the parietal cells and the pepsin-forming chief cells. Schaffer has shown that the former occur in the cardiac glands of man in varying numbers. This observation I can confirm. The parietal cells of the human cardiac glands exhibit the characteristics which have been described for those of the fundus glands by Zimmermann, 98, are frequently binucleated, and contain distinct intracellular ducts. The parietal cells are not confined to any special region of the cardiac mucosa nor to any definite part of the gland, although I have not observed any in relation with the cylindrical epithelium of the foveolæ.

Schaffer also saw among the usual transparent cardiac glands certain more deeply staining tubules which he describes as follows:

“Daneben findet man Drüenschläuche welche nahezu cubische Zellen mit compactem stark gefärbtem Protoplasmakörper und kugeligem Kern besitzen; dies sind offenbar im Gegensätze zu der vorigen durch das Reagens nicht veränderte oder ruhende Zellen vor oder nach der Secretentleerung.” These may have been resting cells, as Schaffer suggests, or zymogenic chief cells, which I have found to be of quite frequent occurrence in my preparations. In Müller's fluid preparations the distinction between these two kinds of cells is not obvious owing to the solvent action of the fluid on the granules and prozymogen.

In my material these zymogenic chief cells occurred even more frequently than parietal cells. Their presence served to throw into strong relief the differences between them and the ordinary cardiac gland cells. Like the parietal cells they showed no special preference for any part of the cardiac region but occurred in the highest as well as the lowest portion of it. In the simple tubular glands of the distal region, they were usually found at the deep end of the tubule, but in the complex tubulo-racemose glands near the œsophageal epithelium their arrangement was less definite. Sometimes they occupied in common with a few parietal cells the terminal tubules of a gland. Some of the large compound glands at the margin of the œsophagus were almost exclusively zymogenic, in others some of the tubules were mucous, the rest zymogenic, while in the majority of cases one or two tubules only or a few cells in some of the tubules presented the structure of chief cells. The tubules formed of zymogenic chief cells are as a rule narrower and have a smaller lumen than those formed of mucous cells.

In sections stained in toluidin blue it is possible to distinguish the two kinds of tubules in the cardiac glands with a very low power, owing

to the intensity with which the prozymogen (basal filaments) in the outer zone of the ferment-forming chief cell stains. Fig. 4 shows a group of such tubules surrounding the ampulla-like duct of one of the compound gland groups at the margin of the œsophagus.

By staining in Heidenhain's iron hæmatoxylin the structure of these cells can be shown to be exactly like that of the chief cell of the body

FIG. 4. Portion of compound cardiac gland from the edge of the œsophagus of man, showing the two kinds of secreting cells of which it is composed. $\times 100$.

of the fundus gland (Fig. 3, *c*). The outer zone of the cell is deeply stained and indistinctly striated owing to the presence of prozymogen. The inner zone contains well-defined zymogen granules which were fortunately fairly well preserved in my material. The zymogen granules were slightly smaller and less closely aggregated than in the chief cells of the fundus glands.

There was no constant relation between the point of occurrence of the parietal cells and the zymogenic chief cells in these glands.

The transition from the cardiac gland zone to the fundus region is a gradual one. As the fundus region is approached chief cells and parietal cells begin to be of frequent occurrence in the deep ends of the glands. These gradually increase in number and occupy more and more of the gland until all but the neck and foveola is formed by them. The change takes place thus by a reduction in number of the mucous cardiac gland cells and a substitution for them of chief cells and parietal cells in the body of the fundus gland. This transition, as well as the relationship of the cardiac gland cells to the epithelium of the gastric foveolæ strongly suggests a relationship of the cardiac gland cells to the mucous chief cells of the neck of the fundus glands and the pyloric gland cells which I have shown to be closely related (see also Cade, 1900). On examination, it is found that they closely resemble both of these elements without being actually identical with either. The neck chief cells and pyloric gland cells stain more intensely in mucicarmine and muchamastein than the cardiac gland cells; the cells are more completely filled with secretion and more uniform in this respect in the fundus and pyloric glands. In the cardiac glands, on the other hand, the cells vary within wide limits as to their degree of physiological loading, but as a rule exhibit a proximal protoplasmic zone of some extent. Still, if one compares the more loaded and deeply-staining cardiac gland cells with the cells from the two other sources, the distinction disappears completely.

In one of his cases, Schaffer made an observation which is of considerable interest if it should prove to be well founded and the clearness of his figures and the confirmations recently furnished by Hari, 01, and Boekelmann, 02, leave little room for doubt. He found among the usual crypts certain others in which well-defined goblet cells could be recognized among the usual cylindrical mucous cells. He also saw cylindrical cells with a well-defined striated cuticle with basal granules and all stages of transition between the latter and mucin-forming gastric epithelial cells. This suggests the possibility, in which, however, Schaffer does not concur, that the resting epithelial cells of the stomach are morphologically identical with the cells of the intestinal epithelium. Schaffer rather prefers to regard the gastric epithelium as specific and the cell types described as dislocated intestinal epithelium. It is difficult to comprehend how this developmental feat could have been accomplished and the mere presence of a striated cuticle is not in itself sufficient to stamp an epithelial cell as intestinal in nature. I have not found any such cells in my preparations, but I have seen some structures which could be readily mistaken for a striated cuticle. The gastric

epithelial cells frequently show a peculiar radial striation in the mucigenous border due to the optical projection into this border of structures which are really on the surface of the cell. Carlier, 99, has shown that in the gastric epithelium of Triton the cement substance is not confined to the contiguous margins of the cells but extends in radiating lines over their free surfaces. This may be particularly well seen in sections of the newt's stomach stained by the iron hæmatoxylin method. Similar features are also exhibited by the cement substance of the gastric epithelium of mammals. In longitudinal sections of the cells in iron hæmatoxylin preparations the cement lines are seen as black dots between the free edges of the cell, or as a line stretching across the cell near its free end, according to the focus. In the latter case fine lines of cement may be traced from the intercellular line in the direction of the end of the cell over the convex surface of the mucigenous border. At the point where these vertical lines join the cement line, there is a granule-like thickening, which might readily be mistaken for the basal granule of a striated cuticle. The same appearances may be made out by careful focussing in a surface view of the ends of the cells where it is seen that the lines of cement over the surface of the cell form an irregular network with nodal thickenings (Fig. 5). This is undoubtedly the explanation of the appearance represented by Schaffer in his Fig. 44, where a row of granules is shown along the free border.

FIG. 5. Surface view of cells of the surface epithelium of the human stomach. $\times 1500$. Stained in iron hæmatoxylin to show cement lines between the cells and the fine lines of cement substance which are continued over the free surface of the cell.

From the foregoing facts it is obvious that the differences between the fundus glands and cardiac glands are not fundamental. This is indicated by the occurrence in the cardiac glands of all the characteristic glandular cells of the fundus glands. Only the relative frequency of the various cells is altered. The cells which are most numerous are muciparous cells morphologically equivalent to the chief cells of the neck of the fundus gland and to the pyloric gland cells, although physiologically much less active than these. In view of the fact that the highly branched glands may be to a large extent formed of zymogenic chief cells which are the morphological equivalents of the chief cells of the body of the fundus gland but little importance can be attached to the mode of branching as a specific character of the cardiac glands.

It now remains to be seen to what extent these conclusions can be supported by data derived from the examination of other mammalian stomachs.

II. THE CARDIAC GLANDS OF THE FIG.

It is in this animal among placental mammals that the cardiac glands reach their greatest extent. They occupy fully one-third of the available surface of the mucous membrane, comprised in a triangular area at the left extremity of the stomach. The mucous membrane of this area is only a fraction of the thickness of that of the middle or fundus gland region and contains throughout glands composed of but one kind of cell.

FIG. 6. Vertical section of tunica mucosa of the cardiac gland zone of pig. $\times 72$.

As regards the general arrangement and shape of the glands and the nature of the interglandular tissue, little can be added to the admirable descriptions of Ellenberger, 88, and Greenwood, 85. Fig. 6 shows a section of the mucous membrane of this region as seen under a low power objective. The superficial portion of the mucous membrane is occupied by the closely-set foveolæ of the glands, which are much narrower and more numerous than in the fundus region. The foveolæ descend in the mucous membrane, becoming narrower as they go and at their lower ends receive one or two glands. These are wavy tubules branching but slightly, usually very narrow at their upper ends but frequently presenting at their lower ends a bulb-like expansion containing a larger lumen.

The surface epithelium of this stomach, while similar in all important details to that of other mammals is unique in one respect; the theca of mucus is of extraordinary size and as a result nearly fills the cell, the nucleus being flattened by compression in the base of the cell. Similar cells line the upper portions of the foveolæ, but it is seen that on going down the sides of the latter the mass of mucus becomes relatively smaller and the nucleus rapidly expands to its normal spherical shape.

The character of the cells lining the deep ends of the foveolæ and the glands differs according to whether the animal is mature or otherwise. Fig. 7 represents the upper end of a cardiac gland and the adjacent portion of its infundibulum from an animal about six weeks old. The preparation was stained with Mayer's muchæmatein followed by acid rubin. The upper cells present two distinct zones; a larger distal zone filled with closely-packed granules of mucigen, black in the figure, which were stained blue by the muchæmatein, and a proximal protoplasmic zone containing the slightly flattened nucleus. Proceeding down the gland it is presently observed that the mass of mucigen exhibits an hour-glass constriction in the center, owing to a concentration of the protoplasm of the cell at that point, and still further down, is completely divided into two masses, one of larger size near the nucleus, and a smaller band along the free border.

On close inspection, the bridge of protoplasm separating these two masses is found, in sublimate preparations to be studded with minute granules which stain intensely in eosin or rubin S. At the lower ends of the foveolæ the proximal or internal mass of mucigen disappears altogether and in the whole of the gland proper only a slightly blue stained band is discernible along the free borders of the cells. As the internal mass of mucigen diminishes in size there is, however, a corresponding increase in the number of rubinophilous granules which occupy, in the upper portions of the glands one-third or more of the cell near the lumen.

These granules are not zymogen granules, because they are not visible in the fresh cell and because Greenwood, —, has shown that the cardiac mucous membrane of the pig yields, when tested by Grützner's method only one-eightieth as much pepsin as the fundus region. The

FIG. 7. Lower end of foveola gastrica and adjacent portion of cardiac gland of the young pig. $\times 1000$. From a preparation stained in strong muchæmatein and rubin. Portions stained blue in muchæmatein are indicated black in figure.

gradual disappearance of the granules as the internal mass of mucigen increases in size points to the probability that the latter is to some extent formed at the expense of the substance of which the granules are composed. Similar granules have been observed after fixation in sublimate solutions, by Carlier, 99, in the epithelial cells of the stomach of Triton, and I have myself observed them in many of the mucous cells of the stomachs of different vertebrates, more particularly in the actively dividing cells at the bottom of the foveolæ. As to their nature, the researches of Krause, 95, on the retrolingual gland of *Erinaceus* seem to be conclusive. He found that at a certain stage of secretion the cells contained no mucin but, in sublimate specimens, were studded with minute rubinophilous granules. These granules were not visible in the fresh salivary gland and their presence was explained by the assumption that the cell juice, preparatory to secretion was rich in dissolved proteids and that these were precipitated in the granular form by the corrosive sublimate employed for fixation.

The narrow tortuous upper portion of the gland in the young pig is entirely composed of cells of this type, exhibiting in such preparations a proximal protoplasmic clear zone containing the spherical nucleus, and a distal zone filled with granules of precipitated proteid. The expanded lower end of the gland is formed of small cubical cells with nearly spherical nuclei (Fig. 8A). The protoplasm of these cells is similar to that of the proximal zone of the cells last described, and may be quite devoid of rubinophilous granules, although a few may usually be seen along the lumen.

Examined for masked iron by Macallum's method, the protoplasm of these cells gives a very feeble reaction, indicating that prozymogen is present only in traces. Treated with muchæmatein or mucicarmine solution, the cells of the lower expanded extremities of the glands of the young pig like those cells which contain numerous rubinophile granules, either remain unstained or show a faintly-stained distal margin as in Fig. 8A, although the cells of pyloric glands of the same animal are filled with mucin in the various stages of its elaboration, and stain intensely in the solutions mentioned. There is thus at this stage apparently very little ground for assuming any relationship between the cardiac and pyloric glands.

The examination of the cardiac glands of the adult animal, however, shows that the failure of the muchæmatein test in the young pig is but an example of delayed and imperfect functional activity.

In the adult animal the cells of the duct and free surface are much of the same character as those of the young pig, and the same gradual

transition may be observed between the cells of the various parts of the gland, but the cells of the gland have altered considerably in character.

FIG. 8A and 8B. Transverse sections of deeper end of cardiac glands: A, from young pig; B, from adult pig. $\times 1000$. From preparations stained in paracarmine and strong muchæmatein. The deeply shaded portions of the cells next the lumen represent the extent of the mucus stain.

The mucus in these cells stains readily in both mucicarmine and muchæmatein, if employed in the manner described for the human cardiac glands.

The cells of the lower ends of the glands are much the same as in the young animal, but always show a considerable amount of mucin in their distal zone (Fig. 8B) which stains strongly in mucicarmine and muchæmatein, although less readily in the latter than the cells of the surface.

Comparing these cells, now, with those of the pyloric glands, the difference although still striking, is not so great as in the young animal, for while the pyloric gland cells are much larger than those of the cardiac glands and are quite filled with mucin or its antecedent substances, they are nevertheless engaged in the same kind of activity as the cardiac

In the upper portion of the gland where are found in the young animal cells with a large distal zone filled with rubinophilous granules, the adult gland contains cells with a distinct mass of mucus. This mass may be confined to the interior of the cell in which case, a protoplasmic portion filled with granules intervenes between it and the free border of the cell, or it may reach the free border in which case the granules are clustered around the proximal end of the mass; or, finally, it may be partially or completely divided into two masses by a strand of granular protoplasm stretching across the cell.

FIG. 8B. Transverse section of deeper end of cardiac gland from adult pig. $\times 1000$. See Fig. 8A.

gland cells and are related to the surface epithelium through a similar perfect transition. The difference is one of degree rather than kind, the cardiac gland cell discharging at a later period of development and with obvious reluctance, the mucin-forming function which the pyloric gland cell assumes early and performs perfectly throughout life.

I have not made an exhaustive comparison of the reactions with synthetic dyes of the cells composing the pyloric and cardiac glands, but so far as I have gone, they have been similar. I should not be surprised, however, if further experiment revealed differences of staining capacity, because it is hardly to be expected that imperfectly functioning secreting cells like those of the cardiac glands would always correspond, in the phase of elaboration to which they carried their stored-up product, with such active elements as the cells of the pyloric glands.

The fundus gland zone of the pig is of great interest owing to the fact that the glands of a very considerable portion of it may be reasonably regarded as intermediate between cardiac glands and true fundus glands. These fundus glands are unique among mammalian gastric glands in the fact, first noted by Greenwood, that the mucous chief cells are not confined to the neck of the gland, but extend into the body and many of the glands may be formed of mucous cells and parietal cells to the exclusion of zymogenic chief cells even in their deepest portions. The ferment-forming chief cells are thus relatively reduced in number.

Thus the cardiac glands of the pig correspond with those of man in their muciparous function, in their relation to the surface epithelium, and in their comparative inactivity in secretion as indicated by the structure of the cell. They differ from those of man in the complete absence of parietal and ferment-forming cells except in the intermediary zone.

III. THE CARDIAC GLANDS OF RODENTIA.

In the guinea-pig, the œsophageal epithelium terminates at the opening of the œsophagus into the stomach in a sort of irregular fringe which bears a superficial resemblance to the Grenzfalte of the *Myomorpha* but on microscopic examination proves to be composed of the corneous layer of the epithelium only.

The cardiac glands in this animal are confined to a narrow zone about one-third of a millimeter in width. The glands of this area are devoid of both parietal and zymogenic chief cells. The cells of which they are composed resemble very closely in structure those of the human cardiac glands although the distal portion which contains secretion

stains much more intensely in muchæmatein and mucicarmine. As in the human subject, the mucigenous distal zones of the cells vary in extent, from a narrow border along the lumen to one-half or two-thirds of the cell. Rarely the whole cell is filled with secretion but not to such an extent that the shape of the nucleus is materially altered by compression.

The transition from this area to the fundus gland area, appears under a low magnification to be an abrupt one, but closer inspection shows that this is due to the sudden appearance of the parietal cells in great numbers. The glands which immediately succeed the cardiac glands are composed entirely of mucous cells and parietal cells. The ferment-forming chief cells make their first appearance at the very bottom of the glands, increasing rapidly in number and displacing the mucous cells until the latter are confined to the neck of the gland. Thus, there is, if one leaves the parietal cells out of consideration, a gradual transition between the cardiac glands and fundus glands. There is also a gradual transition in the nature of the cells composing the gland to the cells of the free surface similar to that shown to exist in the pig and in man.

In *Arctomys monax* the cardiac glands occupy a zone about 2 mm. in width around the opening of the œsophagus. They are for the most part simple tubular structures slightly expanded at the lower extremities, and opening at the upper end into wide and deep depressions. Neither parietal nor zymogenic chief cells are present in these glands and the cells differ from those of the animals already considered in the fact that the secretion completely fills the cell, the nucleus on this account exhibiting a basal position and a crescentic form. The transition to the fundus zone is as in the guinea-pig except that the parietal as well as the chief cells *gradually* increase in number as the fundus gland region is approached. The secretion of the cells stains readily in muchæmatein, mucicarmine and particularly in methyl blue, enabling one to distinguish readily the cells from the ferment-forming cells which in *Arctomys* as in the Rodentia generally, exhibit a very well-developed proximal zone filled with prozymogen.

In the *Myomorpha*, with the exception of *Myoxus*, the stomach is divided into a right and left portion. The glands are entirely confined to the right division of the stomach, the left being lined by a non-glandular stratified squamous epithelium similar to that of the œsophagus. The line of junction of the two divisions is marked internally by a well-defined fringe, the *Grenzfalte*, which is really a fold of the mucous membrane. Corresponding to this externally there is a more or less

obvious constriction. In connection with the formation of this left non-glandular region there is an enlargement of the caecal left extremity of the stomach, in some cases to such an extent that it exceeds in size all the rest of the stomach.

In *Mus musculus*, both the "Grenzfalte" and the external constriction are little marked and the stomach of this form, therefore, represents, with the exception already noted, the simplest type known in the *Myomorpha*.

The extensive cardiac gland zone described by Toepfer, 91, adjoining the opening of the oesophagus on the lesser curvature of the mouse's stomach, did not occur in any of the numerous specimens examined by me. The glands of this region are relatively short, it is true, but they

FIG. 9. Section across the junction of the right and left divisions of the stomach of *Mus musculus*. $\times 500$. Iron haematoxylin.

contain all the characteristic elements of the fundus glands. There are, nevertheless, cardiac glands in a different situation. In a section across the line of junction of the two main divisions of the stomach it is seen that there are along the "Grenzfalte" two or three rows of glands which may, I think, on account of their structure, be properly designated cardiac glands. The epithelial and glandular portions of such a section are represented in Fig. 9. The gland nearest the "Grenzfalte" is usually a simple crypt lined by cells like those of the surface epithelium, but not infrequently there are two or three glandular cells at the lower end of the tubule. These cells exhibit the usual structure of mucous cardiac gland cells and their contents stain readily in mucicarmine and muchæmatein. The simple crypt above described does not occur with the same frequency along the "Grenzfalte" as the other glands, and if serial sections are not examined may be readily overlooked.

The transition between these cells, when present, and the surface epithelium is usually abrupt, but this probably is due to the shortness of the glands. The parietal and ferment-forming chief cells make their appearance in the third or fourth tubule, the former being usually the first to appear.

The limited cardiac gland area extends all along the junction of the two divisions of the stomach, separating the stratified epithelium from the glandular area.

The stomach of the American water-rat or Muskrat, *Fiber Jibethicus*, resembles closely in structure that of *Arvicola arvalis* as described by Retzius, 41, Toepfer, 91, and others. The stratified epithelium which is confined in the mouse to the left division of the stomach, in the muskrat passes over into the right division, replacing the glands of the lesser curvature, ventral and dorsal surfaces and most of the pyloric glands. The fundus glands are confined to a small isolated oval area on the summit of the greater curvature, surrounded on all sides by a non-glandular mucous membrane covered by stratified squamous epithelium which forms a particularly well-marked "Grenzfalte" on all sides.

The pyloric glands are not so completely displaced as in *Arvicola*. A small ring-shaped area still remains around the pylorus, from which a tongue-shaped mass extends along the lesser curvature in the direction of the oesophagus. These pyloric glands are separated from the adjacent cutaneous region of the stomach by a "Grenzfalte" somewhat less developed than that surrounding the islet of fundus glands.

As in the mouse, there is between the "Grenzfalte" and the fundus area on all sides a narrow strip of cardiac glands. These glands have neither parietal cells nor ferment-forming chief cells. The cells composing them contain next the lumen a mass of secretion which stains readily in the usual mucin stains. The secretion does not fill the cells, a large protoplasmic proximal zone containing a spherical nucleus being usually present. Occasionally, as in the human subject, the glands are cystically dilated, in which case the cells are almost wholly protoplasmic.

The pyloric glands of the muskrat are scarcely distinguishable from the cardiac glands, but on the contrary, differ from the glands of Brunner inasmuch as the cells composing the latter are quite filled with mucin, and possess a flattened crescentic or irregular nucleus.

An interesting fact in connection with the stomach of the muskrat is that the fundus glands which are nearest the pylorus exhibit a structure similar to that of the fundus glands of the pig, inasmuch as mucous cells occur not only in the neck of the gland but are frequent among the ferment-forming chief cells of the bottom of the gland.

In addition to the animals included in the preceding description, which were selected because they have an important bearing on the discussion of the phylogenetic significance of the cardiac gland with which this paper will be concluded, I have examined the cardiac glands of a number of other placentals and of one marsupial, for example, the

cat, dog, rabbit, hedgehog and opossum. The results in these instances correspond so closely with those in the animals selected for description that it was not considered desirable to prolong the histological portion of the paper by further description. In every case I was able to satisfy myself that the cardiac gland cells were mucin-forming elements, staining in the modified solutions of Mayer and related in the usual way to the surface epithelium.

In the opossum, the only marsupial at my disposal, there is a narrow zone of cardiac glands, around the cardiac orifice of the stomach. These are tortuous tubules entirely composed of mucous cells which stain readily in muchæmatein and mucicarmine.

It should be remarked that as a rule in those animals in which the cardiac zone is extremely small the cells are relatively more active in secretion than in animals in which the cardiac zone is large.

SUMMARY OF RESULTS IN THE HISTOLOGY OF THE CARDIAC GLANDS.

As regards the nature of the cardiac glands, my results are directly opposed to those of Ellenberger, Edelmann and Schaffer, who concur in the conclusions that they are not mucous glands. Ellenberger contrasts them with the pyloric glands which he regards as mucous glands, and expresses the opinion that they are serous glands. Edelmann regards the cardiac glands as *sui generis*, but not mucus-forming. Schaffer compares them with the pyloric gland cells and the chief cells of the fundus glands which he evidently regards as similar.

Little importance can be attached to any of these views because they are obviously not based on a clear conception of the fundamental structure of the chief cell and pyloric gland cell.

My conclusions are as follows:

1. The cardiac glands are mucous glands, because their cells contain a secretion which stains with Mayer's muchæmatein and mucicarmine and are connected with the mucigenous epithelium of the surface by a transition, the middle point of which is formed by actively dividing cells containing little mucin, which occur at the deeper constricted ends of the foveolar depressions. It is reasonable to suppose that cells which have a common origin from an element that is already differentiated as a secreting cell, are not strikingly different in nature.

2. The cardiac gland cells are fundamentally different from the chief cells of the body of the fundus glands, inasmuch as the latter give none of the staining reactions of mucin, but, on the contrary, contain two characteristic substances which are phases in the elaboration of their

secretion, namely the zymogen granules and the so-called basal filaments containing prozymogen.

3. The cardiac gland cells are closely related to the mucous chief cells of the neck of the fundus gland and to the pyloric gland cells. This conclusion is based on the facts that all three stain similarly in mucin stains, are connected by a similar transition with the surface epithelium, and are recruited from similar dividing cells at the bottoms of the foveolæ. Furthermore, if one traces the transition from the fundus zone to the pyloric zone on the one hand or the cardiac zone on the other, it is found to take place by the gradual lengthening of the neck of the gland, together with the gradual disappearance of the chief and parietal cells.

This similarity does not amount to actual identity of the several structures although such differences as exist are rather of degree than of kind. In the cardiac gland cells as a rule only the distal portion of the cell near the lumen is occupied by the mass of secretion, in the pyloric gland cells and mucous neck cells, it frequently fills the whole cell, the nucleus being compressed and flattened at the proximal end. Moreover, the mucin of the cardiac gland cells stains with less intensity in muc hæmatestin and mucicarmin than that of their prototypes in the other glands. The obvious explanation of these differences is that the cardiac gland cells are physiologically less active and secrete their mucin less rapidly and in a less concentrated form.

4. The peculiar grouping and branching noted by Edelmänn and others in the cardiac glands possess no important significance either as a characteristic of the cardiac glands or as a point of difference between them and the fundus and pyloric glands. The fact that among the human cardiac glands there occur highly complex branched tubular glands, similar in all respects to the other cardiac glands except that they are formed wholly or in part of zymogenic chief cells, indicates clearly that this branching and arrangement are determined by other factors than the phylogenetic history of the glands or the nature of the cells composing them.

5. The cardiac glands are decadent or retrogressive structures derived from fundus glands by the disappearance of their more highly specialized cellular constituents, the zymogenic chief cells and the parietal cells. This conclusion will be discussed at greater length in the next section, but in the meantime the histological arguments in its favor may be stated. These are in brief: (1) the circumstance that both parietal cells and zymogenic chief cells occur in small numbers in the human cardiac glands; (2) the feeble physiological activity of the

cells as indicated by their structure, and (3) the tardy assumption by the cells of the cardiac glands in the pig of the mucigenic function.

THE PHYLOGENETIC SIGNIFICANCE OF THE CARDIAC GLANDS AND THE SO-CALLED CESOPHAGEAL SACS.

Concerning the phylogenetic significance of the cardiac glands the various writers express themselves with considerable reserve and not always with clearness. Edelmann, 89, suggests two possibilities as follows: "Phylogenetisch kann die Cardiadrüsenregion entweder als ein in die Bildung des Magens hineingezogener Abschnitt der Vorderdarmdrüsen oder als ein modifizierter Teil der Schleimhaut des Mitteldarms aufgefasst werden." Between these two possibilities he does not attempt to decide. He, however, regards the cardiac glands as physiologically important structures. "Die physiologische Bedeutung der Cardiadrüsenregion beruht in der Bildung einer Art Vorraum im Magen, welcher keine Säure dagegen Fermentquellen enthält, und in dem die Verdauung der Stärke vor sich gehen kann." "Der von der Cardiadrüsenregion gebildete Vorraum für die Stärkeverdauung kann zum Teil ersetzt werden durch cesophageale Vormägen, so dass die Cardiadrüsenregion also auch mit diesen morphologisch in einem korrespondierenden Verhältnis steht." This last sentence would seem to imply the view that both cardiac gland and cesophageal sac have a common origin, but for the doubt expressed in the first sentence quoted as to the phylogenetic significance of the former.

Oppel, 96, suggested that the conditions described by Schäfer and Williams, 76, Cordier, 90, and others, in the kangaroo, were primitive and that the simple glands or cardiac glands of these animals were of the same nature as the simple gastric gland of lower vertebrates. This suggestion, which, indeed, the author did not seriously advance, may be dismissed with but slight discussion. In the first place, the cardiac glands are not histologically similar to the fundus glands in the lower vertebrates, and in the second place, the more primitive marsupials, such, for example, as the opossum, resemble closely in the structure of their stomachs the simplest conditions found in the placental mammals.

The universal occurrence of cardiac glands in Mammalia seems probable. It is true that their presence in the carnivorous Cetacea and in the ruminants was denied by Edelmann and in three of the suborders of Rodentia by Fleischmann. The latter author, however, did not recognize as cardiac glands the extremely narrow zone of mucous glands found around the cardiac orifice of the stomach in these forms, nor the narrow zone of similar glands found along the "Grenzfalte" in the Myomorpha.

Even if we admit that the cardiac glands occur in all mammals, it does not by any means follow that they have any palingenetic significance.

If we reject the hypothesis of Oppel that the cardiac glands of mammals correspond morphologically to the simple fundus gland of the lower vertebrate classes, the possibilities as to the origin of these peculiar structures become reduced to the following:

1. They are œsophageal glands and the region which they occupy is a portion of the œsophagus;
2. They are modified fundus glands;
3. They are secondary structures of physiological importance which have arisen in the Mammalia in response to a new functional demand.

The first of these hypotheses involves the discussion of the whole question of the participation of the œsophagus in forming the stomach of mammals. It is a well-known fact that in many mammals belonging to different sub-classes and orders, a considerable portion of the stomach is devoid of glands and provided with a stratified epithelium. This is more particularly the case in the Ungulata and Rodentia, but these œsophageal cardiac sacs also occur in the Cetacea, Edentata and Marsupialia and in a few individuals belonging to other orders. The natural conclusion based on the absence of glands and on the character of the epithelium would be that these sacs are produced by a dilatation of the lower end of the œsophagus, and this is the view taken by Ellenberger and Edelmann, and more recently by Kingsley, 99. The latter author in his recent work on Vertebrate Zoölogy expresses his conception of the morphology of these structures as follows:

“Usually the stomach is regarded as the saccular enlargement of the alimentary canal, lying between the œsophagus and the intestine; but when histological and physiological features are taken into account, it is seen that frequently the lower end of the œsophagus expands and takes part in the formation of the gastric enlargement, and that the stomach proper begins only where the gastric glands appear.” This simple hypothesis is not, however, in accord with the results of comparative anatomy and ontogeny, both the older anatomists, as for example, Gegenbaur and Wiedersheim, and the later investigators, Toepfer, Fleischmann, Boas, Cordier and Oppel, agreeing to regard the stomach of mammals as a morphological unit of equal value throughout the group, the cardiac sacs having been produced by gradual modification of the condition found in a simple stomach, such as that of the Carnivora and Insectivora and many individuals of other orders.

Cordier, 90, whose important researches on the comparative anatomy

of the stomach of ruminants entitles his opinion to some weight, expresses himself thus: "On est donc en présence d'une cavité à son maximum de différenciation"—"Je me refuse absolument à croire, par exemple, à l'origine œsophagienne du rumen."

FIGS. 10-16. Diagrams to illustrate the regions occupied in different stomachs by the various kinds of glands.

FIG. 10. Stomachs of: A, Didelphys; B, Dorcopsis; C, Macropus; the two latter after Schäfer and Williams.

Abbreviations: Ca=cardiac glands; F=fundus glands; P=pyloric glands; Oe=portion of stomach without glands and lined by a stratified epithelium.

The alternative hypothesis that the cardiac sacs have been derived by a gradual change in the cardiac portions of the stomach by reason of which the glands have disappeared and the epithelium has changed in character, suggests an examination of the stages, if stages there be, and of the factors that have been operative, in the production of this extraordinary transformation.

Before proceeding with the discussion of this matter, a brief survey will be made of the relations and extents of the various portions of the stomach in a number of representative species of several orders of Mammalia.

In the Monotremata the stomach is lined throughout by stratified squamous epithelium and is quite devoid of glands, although a well-developed group of Brunner's glands is found at the pyloric orifice.

Among the marsupials two types of stomach are met with. In the opossums (Fig. 10, A) Dasyures, Bandicoots and Phalangers the stomach is simple, resembling closely in shape and in the arrangement of its parts those of the insectivorous and carnivorous placentals. A large fundus gland zone occupies the greater portion of the mucous surface, the rest being occupied by the pyloric glands. The cardiac gland zone is either altogether absent

(Phalangista) or occupies an extremely narrow area at the termination of the œsophagus (*Didelphys*, *Dasyurus*, *Perameles*). The œsophageal epithelium does not extend into the stomach.

In the kangaroos, on the other hand, a highly complex condition exists. The stomach is elongated intestiniform in shape and exhibits peculiar colon-like sacculations. The œsophageal epithelium extends some distance into the stomach and a considerable cardiac gland zone is present. As a result of these important modifications, the fundus and pyloric glands are confined to a small portion of the stomach near the pyloric cord. Particularly interesting are the figures of the stomach of *Macropus giganteus* and *Dorcopsis luctuosa*, given by Schäfer and Williams (Fig. 10, B and C). Externally the similarity between these two stomachs is remarkable. The general form, the sacculations along the greater curvature, and the distribution of the fundus and pyloric glands is the same in each, the similarity in shape extending even to the two small secondary cœca at the left extremity of the fundus sac. Some difference exists, however, in the relative extent of the portions lined by œsophageal epithelium and cardiac glands respectively. In *Macropus giganteus* the œsophageal epithelium extends only a short distance from the cardiac orifice, but in *Dorcopsis* it lines the whole of the fundus sac and extends a considerable distance to the right of the œsophagus, the cardiac gland zone being correspondingly reduced. In *Dendrologus bennetti*, according to Beddard, there is also an œsophageal division of the stomach but it occurs in the middle region between the opening of the œsophagus and the pylorus.

A similar diversity of structure exists in several of the placental orders. This has been particularly well investigated by Toepfer for the Rodentia and by Edelmänn, Boas, and especially Cordier, for the Ungulata.

In the former order a simple stomach is found in the Sciuromorpha, Lagomorpha (Fig. 11, A) and Hystricomorpha. In the Myomorpha, on the other hand, there is usually a sac of considerable size lined by squamous epithelium and without glands. The simplest condition is found in *Mus* (Fig. 11, B) in which the glands extend to the œsophageal opening and in which the stomach has retained the simple form. The right and left divisions of the stomach are separated by a moderately well-defined "Grenzfalte" and along this is a narrow zone of cardiac glands. The next phase of specialization is presented by *Cricetus frumentarius* (Fig. 11, C) in which the stratified epithelium extends past the opening of the œsophagus over into the right division of the stomach. The subdivision of the stomach is now clearly indicated externally.

by a constriction. The stratified epithelium is not however confined to the cardiac sac but extends in the form of two wing-like processes preceded by the "Grenzfalte" into the pyloric sac. In *Arvicola amphibius*, according to Toepfer, the stratified epithelium has encroached

FIG. 11. Stomachs of: A, *Lepus*; B, *Mus*; C, *Cricetus*, after Toepfer; D, *Fiber*.

upon the lesser curvature and replaced a considerable portion of its glands.

In *Fiber* (Fig. 11, D) and in a number of species of *Arvicola* this specialization has reached its highest degree, the stratified epithelium extending over into the pyloric sac along the greater curvature, and replacing to a large extent the pyloric glands and the glands of the lesser curvature in such a way that the fundus glands form an islet completely

surrounded by a glandless mucous membrane covered by a stratified squamous epithelium. Around the margin of this islet of fundus glands is a narrow row of cardiac glands and a continuous "Grenzalte."

Although the stomachs of the existing Ungulata have reached a considerable degree of complexity, anatomists have made out an interesting series of gradations culminating in the highly complex ruminant stomach. The simplest conditions are found in the Perissodactyla and in the Pigs among Artiodactyla. In the former the conditions obtaining in the Tapirs, Horses and Rhinoceroses (Fig. 12, A, B and C) are very similar. The stomach is externally undivided. In the mucous membrane four regions are to be distinguished, an œsophageal region lined by stratified squamous epithelium, a cardiac gland region of some width intervening between this and the fundus region and finally a pyloric gland region. The published descriptions are not quite satisfactory as regards the Tapirs but the evidence seems to be that the œsophageal portion is of much less extent than in the horse and rhinoceros, in both of which it forms a sac of considerable size, in the latter fully one-third of the stomach. In the Artiodactyla the simplest condition is found in the Suidæ (Fig. 12, A). Here the condition closely resembles that to be found in the Tapirs but the cardiac gland zone is more extensive.

The next stage of complexity is found in the Peccaries (Fig. 13, B), the stomach of which is divided into three sacs, a middle one lined by stratified epithelium except at the point most remote from the œsophagus where a few cardiac glands are found, a left saccule lined by cardiac glands and a right saccule containing, according to Edelmänn, fundus glands and pyloric glands, according to Cordier pyloric glands only. The stomach of the sheep (Fig. 14, A) is too well known to require any description. The stomachs of the Tragulidæ and Moschidæ differ from those of the better known Ruminants in the absence of a Psalterium and in the less perfectly developed œsophageal groove (Cordier). The stomach of the

FIG. 12. Stomachs of: A, Tapir (after Edelmänn); B, Rhinoceros (after Owen); C, Equus (after Edelmänn).

camel (Fig. 14, B) has been the subject of histological and anatomical study by Pilliet, 85, Boas, 90, and Cordier, 90, whose results harmonize as regards points of fact although the two latter authors differ as to their interpretation of the homologies of the various regions: Pilliet had pointed out that the so-called water cells of the rumen of the camel were lined by a simple epithelium through which glands open. This observation is confirmed by Boas and Cordier, the former of whom

FIG. 13. Stomachs of: A, *Sus* (from Oppel, after Greenwood); B, *Dicotyles* (from Oppel, after Edelmann).

regarded them as the remains of the glands which at one time occupied all the mucous membrane of the rumen. Cordier is the most precise as to the nature of these glands which he describes as follows: "Les glandes découvertes par Brandt étudiées histologiquement par Pilliet, qui tapissent le fond et les parois latérales des poches, sont très courtes, puisque déjà cette région de la paroi est fort mince. Elles ne sont point pourvues de cellules bordantes, et nous ne savons rien sur la nature de leur sécrétion. Il est probable cependant que cette dernière est simplement muqueuse, et il convient peut-être d'assimiler ces régions glandulaires à celles apparemment de même nature, qui avoisinent l'œsophage chez beaucoup d'animaux monogastriques et étudiées dans la série des Mammifères par Edelmann." In brief, he regards the glands of the water cell in the camel as cardiac glands.

The greater portion of cylindrical glandular division of the camel's stomach appears to be occupied by cardiac glands. The mucous membrane of the region is described by Boas as exhibiting marked longitudinal folds, comparable to those of the psalterium of other ruminants. It contains close-set and extremely short gland tubules which Cordier states to be identical with the glands of water-cells. The distal one-sixth of the mucous membrane is occupied by the usual fundus and pyloric glands.

In the Edentata the stomach assumes several peculiar and interesting forms. The nearest approach to the simple form of stomach that is usual to mammals is presented by *Dasytus* (Fig. 15, A), *Myrmecephaga* and allied genera in which the stomach is lined throughout by a simple

epithelium through which ordinary fundus and pyloric glands open. The cardiac glands are restricted in distribution or absent. Specialization is however manifested in the subdivision of the stomach by an external groove into right and left sacs connected on the outside by a broad tendinous band, and in the greatly increased thickness of the wall of the right or pyloric sac. There are no intermediate stages among the Edentata between this condition and the extremely specialized stomachs of *Manis* and *Bradypus* (Fig. 15, B). In *Manis javanica* (Fig. 16), according to Weber, 91, the whole stomach is lined by stratified epithelium. The fundus glands are not, however, entirely destroyed but are confined to a small portion of the greater curvature, where large compound glands which may be interpreted as evaginations of the mucous membrane occur. The cardiac and pyloric glands are represented by highly branched glands occupying the region around the large complex fundus gland above described, the lesser curvature and a portion of the pyloric region near the intestine.

In *Bradypus* (Fig. 15, B) the stomach has an extremely bizarre form. The oesophagus opens into a large chamber subdivided into three parts called by Klinkowström, 95, the cardial stomachs (Cardiamägen), all of which are without glands. Into this opens a long-pointed sac (*b*) the mucous membrane of which is provided with cardiac glands. On the other side the organ is continued into the intestine by a narrow tube, divided into two parts. The first subdivision is occupied, though not completely, by fundus glands. Along the lesser curvature there is a groove lined by stratified epithelium and on the sides of the groove, the squamous epi-

FIG. 14. Stomachs of: A, *Camelus* (modified from Cordier); B, *Sheep* (modified from Oppel).

thelium is separated from the fundus glands by a narrow row of glands called by Klinckowstrom, pyloric glands, but probably correctly interpreted by Oppel as cardiac glands. The final section of the stomach is lined by stratified epithelium and is without glands.

An examination of the schemes presented in the foregoing pages reveals a number of interesting facts, to some of which attention has been already directed by Oppel and others.

FIG. 15. Stomachs of: A, *Dasypus*; B, *Bradypus* (both modified from Klinckowström).

in an organ already highly specialized.

Moreover, in the Ungulata, no existing member of which exhibits a perfectly simple stomach, the nearest approach to this is found in those animals (the pigs in the Artiodactyla, and the tapirs in the Perissodactyla) which have been conservative in other respects, as for example, dentition and foot structure.

The obvious inference from these facts is that the specialized stomachs found in many mammals belonging to different orders, have been differentiated at a comparatively late period and entirely within the

The first fact that strikes one is that in all these orders except the Ungulata, individuals exist, the stomachs of which closely resemble in structure those of many existing Carnivora, Insectivora and Primates.

There are good reasons for supposing that these stomachs represent a persistent primitive condition. They correspond closely in the relations of the different gland areas with the stomachs of lower vertebrates. Furthermore specialization in the stomach of mammals is accompanied by suppression of existing elements (glands, etc.) as well as by increasing complexity of structure, and it is therefore unlikely that primitive conditions would be simulated in a stomach that has been derived secondarily by degenerative or retrogressive processes occurring

limits of the ordinal groups. Any similarity that may exist between various orders in the direction and nature of this differentiation, is probably simply a parallelism due to the operation of similar causes.

Turning now to the question of the origin of the œsophageal divisions of the stomach, we find that there are two possible explanations put forward of which the first is that they are formed by the dilation of the lower end of the œsophagus.

The arguments in favor of this view are in brief as follows:

In nearly all cases where the œsophageal sac is of large dimensions it is traversed from the opening of the œsophagus to the glandular division by a continuous groove called the œsophageal groove. This has been interpreted as the direct continuation of the œsophageal tube, and it has been suggested that the œsophageal sac has been produced by a hernia-like protrusion of that portion of the œsophagus. On this assumption Ellenberger, 88, and Edelmann, 89, include these œsophageal sacs and the œsophagus together in the fore-gut (Vorderdarm), the true glandular stomach, according to these two authors, forming a part of the mid-gut (Mitteldarm). The second

FIG. 16. Stomach of *Manis javanica* (modified from Weber).

reason is based on the supposed specificity of the epithelium, which is in all respects similar to that of the œsophagus. In view of the great plasticity of epithelium of a low grade of specialization, little importance can be attached to the argument because, both pathological histology and normal histology afford many instances of the transformation of a cylindrical or a ciliated epithelium into a stratified epithelium, as a result of the operation of simple mechanical causes, such as friction. Such an instance has been noted by Haycraft and Carlier, 89-90, in the trachea of the cat. Here owing to the peculiar shape of the ring of cartilage, and the mode of insertion of the muscular layer, there is a permanent fold of the mucous membrane at the back of the trachea. As a result of the constant friction between the two layers of epithelium behind this fold the cilia are to a large extent lost and the number of layers of cells greatly increased so as to bear a great resemblance to a stratified squamous epithelium.

As regards the supposed continuation of the œsophagus by the œsophageal groove it is found on examination that the resemblance is but superficial. This has been carefully studied by Cordier, 90, who has shown that the œsophageal groove is produced by change in arrangement of the inner layer of muscle which is contrary to the rule in the œsophagus parallel to the direction of the groove. This layer becomes enormously developed at the two margins to form the lips of the groove and reduced or completely absent in the region corresponding to the bottom of the groove. The development and functional completeness of the groove is proportional to the degree of specialization of the stomach in which it occurs, not the reverse as would be the case if it were a persistent primitive structure. Apart from the inadequacy of the argument advanced in its support, the hypothesis of œsophageal origin is incapable of explaining the facts of structure as we find them.

One of the strongest arguments against this hypothesis is furnished by the stomachs of the monotremes *Ornithorhynchus* and *Echidna* which as indicated above, are lined throughout by stratified squamous epithelium and contain no glands unless indeed a portion of the extensive group of Brunners glands represent a persistent group of pyloric glands. It is only by suppression of the pre-existing glands that such a condition could be reached. That the stomach of the *Echidna* is really a stomach and not a portion of the œsophagus is indicated by the observation of Oppel, 96, that at a period of development at which the epithelium of the œsophagus has already become converted into several layers, that of the stomach is still a simple cylindrical epithelium.

Another example of the stomach which while retaining its simple external form has undergone extensive internal adaptative modifications, is that of *Manis javanica* in which the stratified squamous epithelium is found throughout the stomach although the glands are still retained in parts. By no process of simple addition from the œsophagus could such a stomach be produced.

Equally inadequate is the theory of œsophageal origin, to explain the structure of the stomachs of *Fiber* and certain of the *Arvicolidæ* where the fundus glands form an isolated group surrounded on all sides by stratified epithelium.

It is, of course, at present impossible to definitely exclude the œsophagus from any participation in the formation and development of the cardiac sacs. Until more complete histological information is available concerning the structure of the stomach in the *Cetacea*, some reservation must be made for these forms. But even here it must be remembered that in constructing his hypothetical series to show the phylogeny

of such a stomach as that of *Phocæna*, Weber started from the *a priori* assumption that the first portion was of œsophageal origin. The observation of Jungklaus, 98, that the epithelium of the first chamber of the stomach of *Phocæna* becomes stratified at a very early stage of development is not, in my opinion, sufficient evidence in itself of œsophageal origin.

In all other mammals, the evidence at present available is that the complex stomachs such as those of Ruminants, Rodents, etc., are produced from a simple stomach by progressive specialization in which the œsophagus takes no part, and in which the important phenomena are the suppression of the gastric glands and the replacement of the mucigenous epithelium by a stratified squamous epithelium. Further investigation may show the same to be true of the Cetacea.

It now remains to examine, as far as present information permits, into the cause of the transformation and the stages through which it has passed.

The changes which have taken place in the process of specialization in the stomach may be divided into two categories. To the first belong the changes of structure of the coats of the stomach more particularly of the mucous membrane; to the second, changes of form brought about by the expansion of certain portions of the stomach by the occurrence of constrictions dividing the stomach into successive chambers, or by the formation of saccular outgrowths.

In connection with the first group of changes, namely those affecting the structure of the coats of the stomach, it has been already indicated that the salient features as far as the mucous membrane is concerned are the disappearance of the gastric glands and the replacement of the simple epithelium by stratified squamous epithelium. It remains to be seen by what steps this condition has been reached and what causes have been operative in producing them.

An examination of the stomachs of *Macropus*, *Sus* and *Tapirus* in which the stratified epithelium has advanced but a short distance into the stomach, makes it extremely probable that the process of transformation has begun at the œsophageal opening, has gradually spread over the fundus sac and the left portion of the stomach, finally encroaching in the same gradual manner on the right half of the stomach. A comparison of the stomachs of *Tapirus* and *Rhinoceros*, *Macropus* and *Dorcopsis* is particularly convincing, the main difference being in the relative extent of the non-glandular division which has expanded to include the whole of the dilated fundus of the stomach in both *Rhinoceros* and *Dorcopsis*. In the *Rodentia* again, although, as far as we know,

no recent rodent presents the intermediate stage between the simple stomachs of the squirrels, rabbits and porcupines and the complex stomach of the *Myomorpha*, such a comparison as that of Toepfer shows clearly that the stomach of *Arvicola arvalis* has been produced from such a stomach as that of *Mus* by a continual encroachment of the stratified epithelium on the glandular region proceeding particularly in the sides and lesser curvature, but ultimately including the pyloric gland region. It is furthermore clear from such a series that the lines of least resistance to the encroachment have been afforded by the cardiac glands and the pyloric glands.

It is inconceivable that such an important process should take place without leaving some traces of the stages in the suppression of the glands. In the writer's opinion this intermediate stage is represented by the cardiac glands. It has already been pointed out that in certain mammals, more particularly *Bradypus*, *Dicotyles*, *Camelus*, and *Auchenia*, large areas of cardiac glands occur completely isolated from the other gastric glands and surrounded on all sides by stratified non-glandular epithelium. The supporters of the theory of oesophageal origin would doubtless regard these as oesophageal glands which have been included in the oesophageal hernia by which the sac was formed, but it must be borne in mind that in no case are they covered by oesophageal epithelium, but, on the contrary, have a simple epithelial covering. It is true that Schaffer has described such a condition in his so-called superior cardiac glands of the human oesophagus, but there is no evidence that these isolated structures which have been found in no other mammal, are primitive. The theory of oesophageal origin is inadequate to explain the persistence of these groups of glands.

On the other hand, the theory suggested by Oppel that the cardiac glands form the intermediate stage in the process of complete obliteration of the gastric glands in portions of the stomach affords a simple explanation of the occurrence of these isolated groups of cardiac glands, and is in accord with the results of histological investigation. Some suggestion of this kind apparently occurred to Edelmann, but he was turned aside from the full consideration of it by his failure to find the cardiac glands in the higher ruminants.

The facts in favor of this view are briefly as follows:

1. A layer of cardiac glands is always interposed between the glandless zone with stratified epithelium and the gland-bearing zone. In the further advance of the stratified epithelium into the stomach, the first structures to suffer suppression would be these cardiac glands. As it is unlikely that new glands of a different origin would be produced at

a point where the bulk of the changes are degenerate, a new intermediate zone of cardiac glands can only be produced by the transformation *in situ* of fundus glands.

2. In some mammals, *Bradypus*, *Camelus*, etc., cardiac glands have been retained in the mucous membrane of certain saccular diverticula of the stomach which on account of their shape have not experienced the full effect of the mechanical action of the food.

3. Comparing the stomachs of *Macropus* and *Dorcopsis* a most remarkable resemblance is observed in the external form and in the relative extents of the pyloric gland and fundus gland zones respectively. In *Dorcopsis*, however, the cardiac zone region is much smaller than in *Macropus* and the oesophageal portion much larger. It is evident that this condition has been reached by the suppression of the anterior cardiac glands and their replacement by a stratified epithelium. A similar comparison may be drawn between *Sus* and *Dicotyles*.

4. In a retrogressive process affecting such a structure as the gastric gland composed of several kinds of elements, it is reasonable to suppose that the various elements would disappear in the order of their specialization, that is, by inference, in the inverse order of their power of independent reproduction. Chief cells and parietal cells rarely divide and it is difficult to decide as to their degree of relative specialization. The whole question of the nature of the parietal cells has been reopened by Schaffer's discovery, recently confirmed by Hewlett, **or**, of their occurrence in the glands of the mucous membrane (obere cardialdrüse) of the upper portion of the human oesophagus. It is possible that we shall be forced presently to admit that we know nothing of their nature or function and include them in a common category with those other little understood elements, the crescent cells of mucous glands. Both chief and parietal cells are clearly more highly specialized than the mucous neck cells for the latter sometimes undergo division. In the histological part of the paper attention has been directed to a number of facts which point to the conclusion that the cardiac glands have been so derived from the fundus glands by the disappearance first of the chief cells and second of the parietal cells.

An analysis of the causes that have been operative in producing these changes is beset with unusual difficulties. In his discussion of the results of the research of his pupil, Toepfer, on the stomach of *Rodentia*, Fleischmann, **91**, utters a warning against the too hasty conclusion that the specialization which has taken place in such stomachs as those of the Ruminants and *Myomorpha* is due to an herbivorous diet. He points out that in three out of the four main subdivisions of the Roden-

tia, composed of animals that are as distinctly herbivorous as the Myomorphs, no such specialization has taken place. On the other hand, the fact that in the purely herbivorous Kangaroos, in the leaf-eating sloths, and in the graminivorous and herbivorous Rodents and Ungulates, similar differentiations of the stomach have arisen independently, is surely worthy of serious consideration. Another factor which is discussed by Fleischmann is a possible correlation between dental evolution and gastric structure. He found, however, that in various species of *Arvicola* there was no relation between the complexity of the molar pattern and the structure of the stomach. In any case in the writer's opinion the principle of correlation would not explain, except in a general way, the increasing complexity of the stomach. The direct effect must be exerted through the food. The occurrence of similar specializations in certain Anteaters and in the Cetacea, which subsist on food which is in a sense the chemical antithesis of that of the herbivores renders it necessary to seek the causes of this specialization in conditions which are common to all the animals which possess it. If we conclude that the object of the complex structure of the ruminant stomach is simply to provide increased storage capacity or an antechamber to the stomach in which carbohydrate digestion can go on, then we must seek for wholly different causes in the Cetacea and Anteaters. It seems more probable that similar causes have operated in all the various instances and that chemical composition has been important only inasmuch as it determined the bulk and consistence of the food masses in the stomach. Any discussion of the methods by which these changes have been effected must for the present be largely speculative, and the final solution of the question must rest with the experimental physiologist. There are, however, a number of facts which indicate that the causes have been of a physical, mainly mechanical, nature.

I have already pointed out that in all these stomachs it is probable that the change began at the cardiac orifice and proceeded gradually into the stomach, encroaching more and more upon the normal glandular mucous membrane. This is exactly paralleled according to the experimental researches of Ellenberger, *90*, by the course of the food. According to this investigator, the food does not "circulate" in the stomach of the horse, but accumulates as it is swallowed in the left extremity of the stomach in the fundus sac. When food is again taken that previously swallowed is displaced *en masse* in the direction of the pylorus by the new food which is in turn similarly displaced by the next meal. There is thus a gradual progression of the food from cardia to pylorus. During this process the food is undergoing various changes.

It is being softened by the action of the saliva and of the secretion of the cardiac glands, subjected to the action of bacterial enzymes, and, after it has passed the cardiac zone, to the digestive action of the gastric juice; it is being converted into a softer and more plastic mass. In view of these changes, the fact to which Oppel has called attention that the distribution of the food masses in Ellenberger's diagram corresponds almost exactly with the areal subdivision of the mucous membrane, is not without significance.

The newly-swallowed food is contained in the portion of the stomach which is lined by stratified epithelium, the cardiac gland region is occupied by food which has already undergone some maceration and softening, and the fundus and pyloric gland zone contains food in which the softening process has reached its highest degree. I can see no other explanation for these circumstances than that the changes have been primarily due to the mechanical action of the food on the mucous membrane, and that the rapidity of their production in any region bears a direct relation to the consistency of the food that is contained in that region. This mechanical effect would be in part due to friction owing to the unusual consistence of the food, but doubtless other factors play an equally important part. Among these, over-distention of the stomach, owing to the greatly increased bulk of the food as compared with that of animals such as carnivora which subsist on a more concentrated diet, is of the first importance. This distention is in placental mammals most prominently displayed in the fundus sac, the portion where histological changes first appear. Another factor of importance is the prolonged stay of the food in the stomach owing to the increased bulk, unfavorable consistence and reduced digestibility. A fourth factor which may possibly play a considerable rôle in aquatic animals with a reduced dentition, is the reduction of the temperature in the mucous membrane due to the rapid ingestion of considerable masses of food of lower temperature than the body.

That such conditions may be the cause of profound changes in the mucous membrane of the stomach is indicated by the recent study by Cade, *or*, of the histological changes in the mucous membrane of the cat's stomach due to a gastro-enterostomy. In this experiment the cat's stomach was divided completely into two portions, one containing the fundus zone and the other the pyloric zone, and the cut ends closed by suture. An anastomosis was then effected between the greater curvature and a loop of the ileum. After a lapse of six months the animal was killed and the stomach examined, with the result that the mucous membrane was found to have undergone changes in the neighborhood of

the new outlet of such a nature that the parietal and chief cells had disappeared from the glands and their places had been taken by muciparous elements similar to but not identical with those normally found in the neck of the gland. It might of course be urged that these changes had been of inflammatory origin due to the surgical injury inflicted but it seems more reasonable to suppose that they have been due to the fact that a new focus for the motor activities of the stomach has been set up and that this coincides with the opening into the intestine. If such change can be produced in the life of a single individual by surgical interference, it is not difficult to believe that just such simple causes acting through many generations and assisted by the processes of natural selection have produced the changes which we see in so many herbivores belonging to both the placental and marsupial series.

As to the precise way in which the factors enumerated above have acted, very little can be said because there is as yet no experimental evidence on which to base conclusions. Possibly limitation of the blood supply to the mucous membrane owing to the pressure exerted by the great bulk of food, and exhaustion of the glands from continued over-secretion may be of importance, while actual friction will explain the change in the surface epithelium.

A further argument in favor of the view that the changes in the stomachs of herbivorous mammals owe their origin to mechanical causes aided by natural selection, is afforded by the occurrence in the sloth, in the camel and the llama and in the peccary of saccular diverticula lined by cardiac glands. If the hypothesis that the cardiac glands are the intermediate stage in the disappearance of the glands, be correct, it is easy to understand why they have been retained in these animals. Suppose, for example, that an animal is undergoing a change of habit of such a nature that it is passing from an omnivorous to an herbivorous diet. The effect of this would be, if the above hypothesis is correct, to cause the transformation of the fundus glands in the neighborhood of the cardiac orifice into cardiac glands and finally the disappearance of the latter and the conversion of the epithelium into stratified epithelium. If, however, in the portions of the stomach where these changes were taking place, sacculation occurred, the mucous membrane of such a saccule would be less exposed to the disadvantageous action of the prolonged presence in the stomach of a food mass of firm consistence or of the friction caused by the movement of the latter over the surface of the mucous membrane. It would be less likely to come into contact with the freshly-swallowed food or to be subjected to sudden changes of temperature. The result would be that the degenerative

processes would be delayed in the mucous membrane of such a sacculus, and that its mucosa would still contain fundus glands long after those of the adjacent portion of the stomach had been converted into cardiac glands. Individuals possessing such saccular diverticula would have a higher digestive potential, and thus a greater chance of survival and of propagation than others less favored.

There would thus be for a time a tendency to preserve and perfect by natural selection these sacculi. Ultimately, however, the degenerative processes would extend even to the mucous membrane of the sacculus and their glands would be first converted into cardiac glands and finally disappear.

We are thus furnished with a means of explaining the retention of cardiac glands in the large pointed sac of *Bradypus*, in the first chamber of the stomach of the peccary, and in the water cells of the camel and llama, while they have disappeared from the rumen, reticulum, and omasum, of the higher ruminants. The cells of the reticulum do not contain cardiac glands because they are not primary diverticula of the rumen. Cordier has produced evidence to show that the partition walls between the cells of the reticulum are produced by fusion of rows of vascular papillæ, such as occur in the rumen.

In the anteaters, *Manis*, and *Echidna* and the duck-bill, the mechanical factor has been introduced in all probability by the admixture of small stones, grains of sand, etc., with the food.

The interpretation of the glands of the water-cells of the camel's stomach and of the pointed sac of the stomach of *Bradypus* as cardiac glands which have been retained long after the glands in the walls of the main cavity of the stomach have disappeared, because of the fact that they have been somewhat removed from the direct action, friction, pressure, etc., of the food, suggests a very interesting field of speculation as to the mode of production of the division into chambers of the stomach of ruminants and some allied changes in form of the stomachs of other mammals. The purpose of specialization of this character is usually conceded to be the provision of storage chambers of sufficient capacity to accommodate the greatly increased bulk of food of reduced nutritive value, and in which the preponderating carbohydrate element may be subjected for a longer period to the amylolytic action of the saliva. That the first of these considerations is of the greatest importance in determining the progressive specialization of the herbivorous stomach there can be little doubt. The precise way in which it acts however is extremely obscure. Moreover, leaving out of consideration the question of the prolonging of carbohydrate digestion, it is not clear

what advantage from the mere standpoint of the storage function a chambered stomach possesses over one in which this purpose is subserved by a mere increase in size of the stomach. On this basis alone, it is difficult to understand the occurrence of a highly specialized chambered stomach in forms like *Cricetus* in which there are supplementary storage chambers in the form of capacious cheek pouches. It is possible, on the other hand, that the division of the stomach into two or more successive chambers has been an indirect effect of the increase in bulk of the food and its altered consistence and has been in reality a conservative effort on the part of the organism designed to check the further advance of the degenerative process in the mucous membrane. The evidence seems to indicate that the natural sequence of changes in the stomachs of herbivorous mammals and of certain other forms which subsist on a peculiar form of food, results in the suppression of the gastric glands beginning at the œsophageal orifice and gradually proceeding from left to right, involving more and more of the mucous membrane. Ultimately, if no conservative process intervenes, this would result in the complete suppression of the gastric glands, as has occurred in *Echidna* and *Ornithorhynchus*.

The subdivision of the stomach by constriction into two or more chambers might be interpreted as such a conservative effort, the object of which is to check the advance of the degenerative process and so provide sufficient storage capacity to meet the needs of the animal with the minimum reduction of digestive potential. If it is true that the disappearance of the glands is due to the mechanical action of a food mass of unusual consistence, it is easy to understand how such a constriction would be of use to the animal by confining the food to the first chamber until it had undergone such a change in consistence from maceration and the action of salivary and bacterial enzymes that it was no longer a source of danger to the mucous membrane. If the constriction rapidly reached such a degree that food could only pass over into the second chamber after a considerable degree of softening had been reached, it would prove an effective barrier to the advance of the degenerative process. This appears to have been the case in the *Tragulidæ*. In many cases, however, the constriction has been insufficient to check completely the degenerative process and merely retards it, as in the stomachs of *Arvicola*, *Fiber*, etc. An interesting example of this condition is afforded by *Camelus* and *Auchenia* in which notwithstanding the constriction, the degenerative process has invaded the distal segment of the stomach and converted the glands of a considerable portion into cardiac glands. This doubtless represents, as Boas, 90,

suggests, an earlier condition, common to the Camelidæ and the ancestral forms of the other Ruminants. In the latter the condition has been effectively met by the occurrence of a second constriction marking off the omasum, but in the Camelidæ the degenerative process has advanced until only an extremely narrow zone of fundus glands remains. A similar explanation is suggested for the peculiar tæniated colon-like stomach of the kangaroos, in which the sacculation may have for its object the retarding of the passage of the food mass from left to right in order to give greater time for the change in consistence referred to above.

A difficulty in the way of accepting this explanation of the origin of sacculation in the stomach is presented by the peculiar stomach of *Myoxus* in which a division into two chambers has taken place in the absence of degenerative changes of any kind whatever, and the peculiar tæniated stomachs of certain Primates, *Semnopithecus*, etc., in which there is an œsophageal division, but a very slight development of cardiac glands. It is possible that an investigation of the latter by more discriminative cytological methods may reveal changes in the proximal fundus glands which would justify their interpretation as cardiac glands. Pilliet's researches only indicate that the glands in question contain parietal cells, they do not give any information as to whether the chief cells which accompany these are pepsin-forming or mucin-forming.

In conclusion, it may be remarked that if the hypothesis of Oppel, which is supported in the foregoing pages, that the cardiac glands are modified fundus glands is correct, it will be necessary to substitute a definition of the cardiac glands based on phylogenetic grounds, for the definition given by Edelman. In the case of the human stomach in the cardiac glands of which both parietal cells and ferment-forming chief cells are present in small numbers, the distinction between cardiac glands and fundus glands might be dispensed with altogether. In the interests of comparative anatomy and particularly on account of the great extent of these glands in the pig, camel, peccary, etc., it seems desirable to retain the present classification and to define the cardiac glands as modified fundus glands occurring at the cardiac orifice of the stomach, or at the junction of the glandular and non-glandular divisions of the stomach, or in special sacs, differing from the usual fundus glands in the reduction in numbers, or complete absence of the parietal or ferment-forming chief cells, or both, and by the increased number of mucous chief cells.

BIBLIOGRAPHY.

- BENSLEY, R. R., 96.—The histology and physiology of the gastric glands. *Proc. Canad. Inst., Toronto*, 1896, vol. i, pp. 11-16.
- BENSLEY, R. R., 98.—The structure of the mammalian gastric glands. *Quart. J. Micr. Sc., Lond.*, 1898, n. s., vol. xli, pp. 361-389.
- BOAS, J. E. V., 90.—Zur Morphologie des Magens der Cameliden und der Traguliden. *Morphol. Jahrb., Leipz.*, 1890, Bd. xviii, S. 494-524.
- BOECKELMAN, W. A., 02.—Untersuchungen zur pathologischen Anatomie des menschlichen Magens in Fällen von Ulcus und Carcinoma bei bekanntes chemischen und motorischen Funktion. *Ztschr. f. klin. Med., Berl.*, 1902, Bd. xlv, S. 128.
- BONNET, R., 91.—Grundriss der Entwicklungsgeschichte der Haussäugethiere. Berlin, 1891. Quoted from Oppel, l. c.
- CADE, A., 01.—Études de la constitution histologique normale, et de quelques variations fonctionnelles et experimentelles des éléments sécréteurs des glandes gastriques du fond. *Arch. d'anat. micr., Par.*, 1901, t. iv, pp. 1-86.
- CARLIER, E. W., 99.—Changes that occur in some cells of the newt's stomach during digestion. *La cellule, Louvain*, 1899, t. xvi, pp. 403-464.
- COBELLI, R., 66.—Le ghiandole acinose del cardia. *Sitzungsb. d. k. Akad. d. Wissensch., Math.-naturw., Cl., Wien*, 1866, Bd. liii, pp. 250-255.
- CORDIER, J. A., 93.—Recherches sur l'anatomie comparée de l'estomac des ruminants. *Ann. d. sc. nat., Par.* 1893. Sér. 7, t. xvi, pp. 1-128.
- EDELMANN, R., 89.—Vergleichend anatomische und physiologische Untersuchungen über eine besondere Region der Magenschleimhaut (Cardiadrüsenregion) bei den Säugethieren. *Deutsche Ztschr. f. Thiermed., Leipz.*, 1889-90, Bd. xv, S. 165-214.
- ELLENBERGER, W., 84.—Handbuch der vergleichenden Histologie und Physiologie der Haussäugethiere. Bd. i, Histologie der Haussäugethiere, Berl., 1884.
- ELLENBERGER, W., 90.—Handbuch der vergleichenden Histologie und Physiologie der Haussäugethiere. Bd. ii, Physiologie, Berl., 1890.
- ELLENBERGER, W. UND HOFMEISTER, V., 85.—Der Magensaft und die Histologie der Magenschleimhaut der Schweine. *Arch. f. wissensch. u. prakt. Thierh., Berl.*, 1885, Bd. xi, S. 249-268.
- FLEISCHMANN, A., 91.—Bemerkungen über den Magen der Rodentia. *Morphol. Jahrb., Leipz.*, 1891, Bd. xvii, S. 408-416.
- GREENWOOD, M., 85.—Observations of the gastric glands of the pig. *J. Physiol., Lond.*, 1884-5, vol. v, pp. 362-372.
- GROTE, R., 97.—Beiträge zur Entwicklung des Wiederkäuermagens. *Ztschr. f. Naturw., Leipz.*, 1897, Bd. lxix, S. 387-479.
- HAMMARSTEN, O., 85.—Studien über Mucin und mucinähnliche Substanzen. *Arch. f. d. ges. Physiol., Bonn*, 1885, Bd. xxxvi, S. 373-456.

- HÁRI, P., 01.—Ueber das normale Oberflächen-Epithel des Magens und über Vorkommen von Randsaumepithelien und Becherzellen in der menschlichen Magenschleimhaut. Arch. f. mikr. Anat., Bonn, 1901, Bd. lviii, S. 685-726.
- HAYCRAFT, J. B. AND CARLIER, E. W., 89-90.—Note on the transformation of ciliated into stratified epithelium as a result of the application of friction. Quart. J. Micr. Sc., Lond., 1889-90, n. s., vol. xxx, pp. 519-522.
- HEWLETT, A. W., 01.—The superficial glands of the oesophagus. J. Exper. M., N. Y., 1901, vol. v, pp. 319-331.
- HUPPERT, K. H., 96.—Ueber die Erhaltung der Arteigenschaften. Prag, 1896.
- JUNGCLAUS, F., 98.—Der Magen der Cetaceen. Jenaische Ztschr. f. Naturw., Jena, 1898, Bd. xxxii, S. 1-94.
- KINGSLEY, J. S., 99.—Text-Book of Vertebrate Zoology, N. Y., 1899.
- KRAUSE, R., 95.—Zur Histologie der Speicheldrüsen. Arch. f. mikr. Anat., Bonn, 1895, Bd. xlv, S. 93-133.
- KLINOWSTRÖM, A., 95.—Zur Anatomie der Edentaten. Zool. Jahrb., Leipz., 1895. Abt. f. Anat., Bd. viii, S. 481-499.
- KUPFFER, C., 83.—Epithel und Drüsen des menschlichen Magens. München, 1883, pp. 1-22.
- LANGLEY, J. N., 84.—On the structure of secretory cells and on the changes which take place in them during secretion. Internat. Monatschr. f. Anat. u. Histol., Leipz., 1884, Bd. i, S. 69-75.
- MACCALLUM, A. B., 98.—On the detection and localization of phosphorus in animal and vegetable tissues. Proc. Roy. Soc., Lond., 1898, vol. lxiii, pp. 467-479.
- MAXIMOW, A., 01.—Beiträge zur Histologie und Physiologie der Speicheldrüsen. Arch. f. mikr. Anat., Bonn, 1901, Bd. lviii, S. 1-134.
- MAYER, P., 97.—Ueber Schleimfärbung. Mitth. a. d. Zool. Station zu Neapel, Leipz., 1897, Bd. xii, S. 303-330.
- NEGRINI, F., 86.—Intorno allo sviluppo e struttura della mucosa gastrica del majale. Gior. di anat. fisiol. e patol. d. animali, Pisa, 1886, v. xviii, pp. 121-148.
- OPPEL, A., 96.—Lehrbuch der vergleichenden mikroskopischen Anatomie der Wirbelthiere. Theil i, Der Magen, Jena, 1896.
- OPPEL, A., 98.—Verdauungsapparat. Ergebn. d. Anat. u. Entwicklungsgesch., Wiesb., 1898, Bd. vii, pp. 29-102.
- PILLIET, A., 85.—Structures des alvéoles de la portion gaufrée de l'estomac du chameau. Bull. Soc. Zool. de la France, Par., 1885, Année x, pp. 40-41.
- RETZIUS, A., 41.—Ueber den Bau des Magens bei den in Schweden vorkommenden Wühlmäusen. Arch. f. Anat., Physiol. u. wissensch. Med., Berl., 1841, Bd. viii, S. 403-420.

- SCHÄFER, E. A. AND WILLIAMS, H., 76.—On the structure of the mucous membrane of the stomach in the kangaroos. *Proc. Zool. Soc., Lond.*, 1876, Pt. i, pp. 165-177.
- SCHAFER, J., 97.—Beiträge zur Histologie menschlicher Organe. *Sitzungsab. d. k. Akad. d. Wissensch., Math.-naturw., Cl.*, Bd. cvi, Abth. iii, S. 353-455.
- TÖPFER, K., 91.—Die Morphologie des Magens der Rodentia. *Morphol. Jahrb., Leipz.*, 1891, Bd. xvii, S. 380-407.
- WEBER, M., 98.—Anatomisches über Cetaceen. *Morphol. Jahrb., Leipz.*, 1888, Bd. xiii, S. 637-653.
- ZIMMERMANN, A. UND SAL, J., 94.—Beitrag zur Histologie des Pansens. *Deutsche Ztschr. f. Thiermed., Leipz.*, 1894, Bd. xx, S. 283-289.
- ZIMMERMANN, K. W., 98.—Beiträge zur Kenntniss einiger Drüsen und Epithelien. *Arch. f. mikr. Anat., Bonn*, 1898, Bd. lii, S. 546-551.