

OBSERVATIONS UPON THE STRUCTURE AND NORMAL CONTENTS OF THE DUCTUS AND SACCUS ENDOLYMPHATICUS IN THE GUINEA-PIG (*CAVIA COBAYA*)

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TWELVE FIGURES

INTRODUCTION

At the suggestion of, and in collaboration with, Dr. G. Carl Huber, director of the department, there is being undertaken in this laboratory a comprehensive study of the circulation of the endolymph from embryological, comparative anatomical, and experimental physiological standpoints. This separate communication on the normal morphology of the ductus and saccus endolymphaticus has become necessary because of the incomplete character of the recorded observations concerning these parts which have been found to be so important functionally in our experimental studies.

It has been customary to designate that part of the membranous labyrinth within the bony aquaeductus vestibuli as the ductus endolymphaticus and that part protruding beyond the external or cranial aperture of the aqueduct as the saccus endolymphaticus. On the basis of both the morphology and the relations to be described, certain modifications of the nomenclature for these parts in the guinea-pig will be suggested. They may or may not prove applicable to the parts in other forms when these have been studied in as great detail, but there is some reason to think that they will.

HISTORICAL

Since other parts of the membranous labyrinth are similar in essential structure in all mammalia and since statements concerning the details of the structure of the ductus and saccus endolymphaticus of the guinea-pig are lacking except for Alexander's ('00) observations upon stages to 16.5-mm. embryos and the brief account of the adult by Portman ('19), it has seemed necessary to review what little is recorded of the histologic structure of these parts in all adult mammalian forms. Such a compilation has not been made in recent years and is therefore of value in itself.

Under the name of 'aquaeductus vestibuli' and its 'sack-like appendage,' Boettcher ('69) described the essential developmental and adult relations and structure of the parts to which Hasse and his pupils ('73) later gave the names ductus and saccus endolymphaticus, by which terms they are now usually designated. Boettcher's excellent work, with its firm morphologic basis, settled forever the dispute concerning the essential nature of the soft tissues in the so-called bony aquaeductus vestibuli—a problem which had vexed anatomists for nearly a century following Cotugno's (1760) report of a sac under the dura from which mercury flowed into the labyrinth—and Boettcher has universally received credit for this demonstration as well as for the first good account of the development of many other parts of the labyrinth. But some of the histologic details given in the shorter 1869 article, based upon free-hand razor sections of adult cat and newborn human material, have not received the attention which it now appears they deserved. He described here the funnel-shaped expansion of the narrow part of the membranous aquaeductus vestibuli into the sac-like appendage, and both described and figured irregular vascularized projections and crypts of the distal part of the aquaeductus and of the adjacent area of the saccus, and further stated in the text matter that the narrower part had a squamous epithelium, the expanding part and the vascularized irregular area a cubic epithelium with more numerous nuclei than the other parts, and that

the distal part of the sac again had a squamous epithelium. This description applied directly to the cat, and he stated that the epithelium in the human newborn was 'flacher' than that of the cat.

That very little attention has been paid to Boettcher's observations of vascularized papillae and of crypts and even less to his mention of epithelial differences becomes evident upon examining the larger works which are even at the present time most consulted for the structure of the membranous labyrinth of mammals, and upon which, either directly or through texts based upon them, the usually accepted views of most anatomists and clinicians as to the structure of the walls of the ductus and saccus endolymphaticus are founded. First place among these very properly belongs to the second volume of Retzius' "Das Gehörorgan der Wirbelthiere," published in 1884. In his review of literature Retzius gives a very complete abstract (pp. 234 and 235) of Boettcher's observations on the ductus and saccus endolymphaticus; but in the descriptions of his own work the most extensive account for any mammalian form is that of the human and consists of the following statements other than those describing location and relations: "Das Epithel des Ductus endolymphaticus ist dem des Sacculus ähnlich. Was das Verhalten des Epithels und der übrigen Wand desjenigen Theils vom Ductus endolymphaticus anbetrifft, welcher durch die Pars petrosa verläuft, so verweise ich auf die eingehende Darstellung von Boettcher; in Betreff der Epithelbekleidung und des übrigen Verhaltens des Saccus endolymphaticus beim erwachsenen Menschen verweise ich auf die von Axel Key und mir gegebene Beschreibung und die Abbildungen" (Bd. 2, p. 341). The Key and Retzius ('75) article referred to describes (p. 215) for the saccus endolymphaticus a low polygonal epithelium, and a surface view is presented in figure 7 of Tafel XXXVII; the authors further state that the ductus has epithelium like that of the saccus. Schwalbe ('87), in his much-quoted "Anatomie des Ohres," says (p. 401): "Das Epithel des Ductus und Sac-

cus endolymphaticus ist ein niedriges Pflasterepithel mit polygonalen Zellengrenzen bei Flächenansicht." In the same paragraph he describes Boettcher's observations with regard to vascularized projections and crypts, but adds no observations of his own. Siebenmann ('97), in the account of the internal ear in the v. Bardeleben Handbuch der Anatomie, makes no definite statement concerning the histologic structure of the walls of these parts. Cannieu ('04), in the chapter on the internal ear in the Poirier and Charpy *Traité d'Anatomie humaine*, calls attention (p. 1346 and p. 1393) to the absence of any neuroepithelium in the saccus, and then says Rauber thought that the pigmentation of certain groups of cells might indicate a vestigial region of nerve termination. No positive statement of structure is made, however. Schäfer ('09), in the eleventh edition of Quain's *Anatomy*, volume 3, part 2, simply says that the parts are lined with epithelium and does not even mention crypts or variations in the epithelium.

Even in Alexander, Marburg and Brunner's recent "Handbuch der Neurologie des Ohres" ('23, '24), W. Kolmer, who wrote the section on the microscopic anatomy of the labyrinth, devotes only one paragraph of fifteen lines to the whole of the ductus and saccus endolymphaticus, and six lines of this are about the selachians and most of the rest about gross relations. He makes the usual statement that it is lined with epithelium, but makes no mention of the structure of this tissue. The only part of real interest to us is the closing sentence (p. 119): "In den embryonalen Stadien zeigt der Saccus endolymphaticus eine zottenartige Entwicklung seiner Wandauskleidung (Tafel V, Fig. 3)." This figure is a poor photomicrograph of a transverse section of some unspecified level of the saccus in a 33-mm. guinea-pig foetus. Kolmer also expresses the very common view that this apparatus "erscheint bei den höheren Wirbeltieren eher als ein rudimentäres Organ; immerhin mag er einen Druckausgleich gegen die Schädelhöhle hin vermitteln." The observations and experiments which form the basis of this and a succeed-

ing paper, and which were reported briefly at the 1924 and 1925 meetings of the American Association of Anatomists, show these views, both structural and functional, to be very much in error; Kolmer's statements merely reflect the lack of knowledge of this part which is common even among the specialists on internal ear.

In view of these very meager accounts in the larger works, it is not to be wondered at that in the more commonly used text-books of anatomy and of otology the statements concerning the structure of the ductus and saccus endolymphaticus are only of the most general nature, and not only incomplete, but also incorrect. It seems probable that this condition is in some measure partially responsible for the lack of real advancement in knowledge of this region; these modern accounts are not as good as the original one given by Boettcher.

Of the very extensive literature concerning development, variations in relations and size, questions of homology, etc., which has grown up since the work of Boettcher, reference will be made only to those accounts in which I have found mention of observations of histologic structure of the walls of the ductus or saccus endolymphaticus in stages later than the embryonic. Accounts in which the statements appear to be in reality merely a repetition of the usually accepted views of the period will not be included.

Hasse ('73) advanced the view that the saccus did not end blindly, but communicated by means of a small opening with the subarachnoid space. He says (pp. 792 and 793): "Ich habe nach den Untersuchungen an Schweine- und Rindsembryonen, deren Gehörkapsel noch knorplig sind und an neugeborenen Menschen, Grund zur Annahme, dass durch eine feine Oeffnung der *dura* ein feiner, trichterförmig sich ausbreitender Fortsatz des an der *apertura aquaeductus vestibuli* gelegenen *saccus* hindurchtritt, mit der *arachnoidea* verschmilzt und somit, wie bei den Vögeln, eine Communication zwischen dem *cavum endolymphaticum* und dem äusseren *cavum epicerebrale (subarachnoideale)* im Gegensatz zum

inneren zwischen *pia* und Gehirn zu stande kommt." Both in birds and in *Siredon pisceformis* Hasse has recorded in this same publication quite large communications of the saccus endolymphaticus with the epicerebral space, and it seems to have been partly in hopes of homologizing all forms that he persuaded himself of the existence of such openings in mammals. By 1881, Hasse himself had very definitely abandoned the idea because of the lack of confirmatory observations. The work of Key and Retzius ('75) and of Retzius ('84) has already been considered above. Rüdinger ('77) gave figures of four sections (out of nineteen serial ones on which he based his description) across an adult human aquaeductus vestibuli, and both showed in his illustrations and stated that the epithelium is simple squamous; one figure shows two folds and another figure some vessels, but Rüdinger said (p. 221) that he could not find the "von Boettcher bei der Katze beobachteten Gefässträubchen obschon man zuweilen verschieden geformten Vorsprünge an der Innenfläche des Sackes begegnet." In a later article Rüdinger ('87) revived Hasse's abandoned hypothesis of openings in the walls of the saccus endolymphaticus. The material used is not definitely stated, but appears to have been entirely embryonic or foetal of cat, dog, pig, beef, and one three-month human. The figures are mostly from the cat, with one from pig and one from human. He described and figured large endothelial-lined spaces alongside the endolymphatic sac and especially surrounding certain 'accessory canals' which he stated to be constant in position and which he regarded as not identical with the processes of Boettcher's descriptions, in that they are from the 'fundus' of the sac. In various parts of the article he called the endothelial-lined cavities 'Lymphspalten' and 'Lymphraumen,' and in one legend of a figure, 'Ductus perilymphaticus.' He stated that the endolymphatic sac communicates with these surrounding spaces both through openings in the ends of the accessory canals ('Abflusskanäle') and through 'scharf abgegrenzte Lücken' between epithelial cells of the most posterior part

of the fundus of the saccus, and that at both regions the epithelium and endothelium form a continuous lining membrane. No figures are given of the communications in the fundus region and but one drawing of a connection from an accessory canal, from a three-month human preparation. Much space is devoted to his functional and comparative anatomical interpretations which will be considered in later papers. It may be stated here that my guinea-pig material shows nothing of such openings. It is difficult, from a study of his figures and a knowledge of the difficulties of histologic technique in this region, to avoid the conclusion that Rüdinger failed to differentiate artefacts due to shrinkage and tearing from the actual structure.

Alexander ('04), in his monograph on the development of the labyrinth in *Echidna aculeata*, says (p. 54), concerning three 'poorly preserved' adult specimens that the saccus has numerous foldings which include the underlying connective tissue and that the saccus is lined by a flattened epithelium ranging from 2 to 8 μ in thickness, while the epithelium of the ductus is from 2 to 4 μ thick. Concerning a 'pouch young' with a head length of 42 mm., he states that the epithelium over the folds, which do not then include the connective tissue, is of a cubic type 7 μ thick.

Sterzi ('09) described the saccus of the newborn and adult human as composed not of squamous, but rather of low cubic epithelium resting on a layer of delicate but dense fibrous tissue about 5 μ thick, outside of which is a loose fibrous 'perisaccular tissue' with numerous blood and lymphatic vessels. An 'uninterrupted' membrana propria is mentioned. Epithelial crests at the borders of the saccus with irregular extensions outward and epithelial trabeculae within the saccus are described; true crypts extending from the infundibular part were observed in only three out of ten adult labyrinths examined for this purpose, but are said to be constant in the newborn. All extensions were observed to end blindly, and direct communications with any of the surrounding vessels or spaces were not found, although

searched for. 'Islands' of epithelium were observed by Sterzi near the fundus of the saccus and between the layers of the dura; these were stated to be distinct from the prolongations of the epithelial crest. In 1919, Siebenmann published an account of the human saccus and ductus endolymphaticus based upon the study of serial sections of twenty-five specimens from cases ranging in age from infancy to seventy-eight years, and including one three-month embryo. Except in one case, a four-and-one-half-month-old infant, he found the epithelium of the ductus and saccus not squamous in type, but "wie es Boettcher beschrieben hat, meistens ziemlich höher als das sog. indifferente Labyrinthepithel, und besitzt in der Regel einen kugligen, grossen, sich intensiv färbenden Kern." He does not mention differences in character in the various parts, as Boettcher did, and his figures do not show any such difference. He adds, however, the very important observation that this epithelium "sitzt, ohne dass sich eine Basalmembran dazwischen schieben würde, den begrenzenden, unter sich parallel verlaufenden fibrösen Dura-fasern oder dem lockeren Zellgewebe direkt auf (Fig. 11 b u. 12 b)." He does not limit this absence of a basal membrane to any particular part, but it is interesting to note that the areas selected to illustrate the points are both from the region of the vascularized processes, concerning the presence of which he, however, states that "nur für ganz seltene Ausnahmefälle" can he confirm Boettcher and others. However, he not only confirms, but extends Boettcher's observations regarding crypts, saying (p. 60): ". . . das betreffende epitheliale Rohr nicht nur reichliche Faltenbildung und seitliche lange enge Ausstülpungen zeigt, sondern . . . es im höheren Alter, namentlich in seinem oberen und mittleren, häufig auch in seinem unteren Abschnitt aus einem ganzen Konvolut von drüsenartigen feinen Kanälchen besteht." He finds no openings from the saccus into any structures in the surrounding loose areolar tissue, which he records to be especially marked in the region of these processes; and he definitely states (p. 61) that 'dies lockere Maschengewebe' is not

in connection with the perilymphatic space, since in the region of the apertura interna of the aquaeductus are found "nurd erb fibröse solide Wandungen." He says that the 'unterste Abschnitt' of the saccus has a simple narrow lumen in the midst of dense dura, often showing neither folds, papillae, nor crypt formation, at least in the newborn and in children. He conceives these formations of older age to be due to 'Involutionenprozessen' and to be at the expense of the original lumen. No mention is made by Siebenmann of the work of Sterzi.

Portman's ('19) brief account of the relations and structure of the sacculus, ductus, and saccus endolymphaticus in the adult guinea-pig is based on serial sections; the only parts of the technique mentioned are immersion fixation, celloidin embedding, and serial sectioning, and no mention is made of fluids or stains used or of the thickness of the sections. A very diagrammatic representation of the gross anatomical relationships is the only illustration given. He says that the saccus endolymphaticus has a wall composed of a simple cubic epithelium with a basement membrane and of connective tissue. The epithelium is said to be higher 'externally' and more flattened as the ductus is approached, and some differences are noted in the amount of connective tissue in regions. The wall is apparently considered to be wrinkled rather than to have true processes. The epithelium of the ductus is reported as simple cubic toward the saccus and flattened toward the junction with the sacculus. Portman also states that there are loosely joined connective-tissue spaces about the saccus and ductus which are continuous with the perilymphatic space. In the discussion of my observations, the papers of Sterzi, Siebenmann, and Portman will be further considered.

It is to be regretted that the excellent studies by Eichler, Siebenmann, Shambaugh, and their pupils on the vascularization of the labyrinth have not included more concerning the details of the vascular network around the ductus and saccus endolymphaticus. In the human Siebenmann ('94) stated

that the tissues of the aquaeductus vestibuli are supplied by a branch (not included in the illustrations) of the posterior vestibular artery, and Nabeya ('23) mentions and illustrates a capillary network about the ductus. Shambaugh ('03) shows a somewhat richer network for the pig, while for the guinea-pig Nabeya ('23) states that he could not find any such network. A great deal of attention has been devoted to the variations in formation of the vena aquaeductus vestibuli otherwise. The saccus endolymphaticus is not shown in the illustrations of any of these authors or of others who have given accounts of the vascularization of the adult labyrinth, and in the case of the guinea-pig, at least, I am sure that if Nabeya's material had included even the whole length of what is usually termed ductus he would have found a vascular network which is absent, as I will later show, in the part which he did figure. Portman ('19), for the guinea-pig, mentions a vascular network which drains by a large stem into the lateral sinus, and also mentions the usual vena aquaeductus vestibuli; he was using only sections of uninjected material, and no mention is made of the extent of the areas drained in each direction. Sterzi ('09) says that when injections are made through the dura with a capillary pipette into the 'perisaccular tissue' the vessels around the saccus in the human can be seen to empty into the transverse sinus. Streeter's ('16) study of these vessels in human embryos extends only to the 130-mm. stage, and his figure 5 of this stage shows only the larger stems, between which he states there are numerous 'small anastomosing capillaries' covering the whole appendage, and "richer over the sac and over the proximal flaring portion of the ductus" and "more scant over the narrow portion of the duct."

These vascularization studies need to be extended to include the relative richness of blood supply to the various parts of the saccus and ductus at all stages for several forms, and it is planned to carry out such studies in this laboratory in connection with the general problem of the circulation of the endolymph.

MATERIAL AND METHODS

These observations have been made upon material prepared by two methods. First, by the method used in the author's previous work upon the labyrinth of guinea-pigs and reported in the 1919 article, to which the reader is referred for the detailed technique (Guild, '19). It is the method with which, after testing many procedures, I have had the best results in cochlear preparations and in brief consists of fixation by vascular injection of a warm Zenker-formalin fluid following warm saline solution, celloidin embedding with special precautions followed by 'double' embedding in paraffin, serial sections at 7 or 10 μ (less if desired), and stained with Heidenhain's iron-lac hematoxylin with a counterstain of benzopurpurin. Secondly, the material prepared in the course of the study of the circulation of the endolymph, with a technique designed to show up the location of the Prussian-blue granules, has also been used in this study. The technique of this method will be given in the account of the experimental work. While not well adapted to the study of histologic detail, it has furnished a sufficient number of series to permit of the study of individual variations in animals of the relationships to other parts.

All of the series by both methods were cut approximately lengthwise of the ductus endolymphaticus; in eleven of them I was fortunate enough to so orient the block that the whole length of the lumen of the ductus, together with adjacent parts of the sacculus and of the saccus endolymphaticus, was included in one section. Two of these cases are shown in outline in figures 1 and 2. These eleven series have made possible more accurate determinations of certain dimensions than could have been done otherwise without resort to reconstruction methods. Because of the number so favorably oriented and the difficulties of making proper allowances in obliquely oriented series, the measurements given of various parts are based entirely on these eleven series.

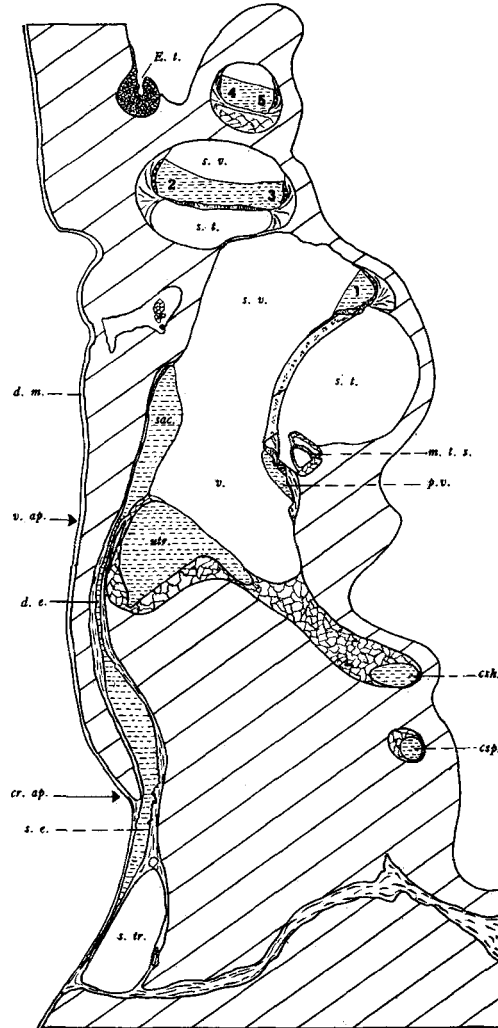


Fig. 1 Outline drawing of a single section of the internal ear of a guinea-pig. Guinea-pig A 22, left ear, slide 5, row 3, section 4. $\times 15$. The plane of the section is approximately horizontal and is parallel and superior to the modiolar axis of the cochlea. The entire length of the lumen of the ductus endolymphaticus is included in this single section; the rest of the section has been sketched, in order to show the orientation with reference to other parts. The narrow part of the lumen of the endolymphatic duct has been slightly enlarged so as to show it at this magnification, but otherwise the proportions are exact. Bone is represented by the oblique hatching. For emphasis, all parts of the lumen of the membranous labyrinth have been indicated by a fine broken horizontal hatching.

No age data were available for the animals used; they ranged in weight from 175 grams to 450 grams, and the majority were in the heavier group. In all cases the animals were in good health.

The drawings have all been made by the author, and those showing details of structure are from a series in which the fixation is especially good. In both outline and detail figures bone has been indicated simply by oblique hatching.

All measurements were made with a Zeiss ocular screw micrometer, using objectives suited to the distances concerned. The determinations of epithelial thicknesses and nuclear dimensions are all based on material prepared with Zenker-formalin fixation. No allowances for shrinkage have been attempted.

RELATIONS AND STRUCTURE

Figures 1 and 2 are outline drawings of single sections from two of the eleven series mentioned in the account of material and methods which were so oriented that one section includes the entire length of the lumen of the ductus endolymphaticus. They are reproduced at a magnification of fifteen diameters; at this scale it was necessary to show the narrowest part of the ductus slightly larger than it really is in order to prevent the lines blending in the engraving process, otherwise the proportions are exact. The lumina of all parts of the membranous labyrinth are shown with a broken horizontal hatching.

The plane of these sections passing through the ductus is approximately parallel to and somewhat more than a milli-

cr.ap., *v.ap.*, the arrows indicate, respectively, the cranial (or external) and the vestibular (or internal) apertures of the osseous aquaeductus vestibuli; *csb*, horizontal semicircular canal; *esp.*, posterior semicircular canal; *d.e.*, ductus endolymphaticus; *d.m.*, dura mater; *Et.*, eustachian tube as it opens into middle ear; *m.t.s.*, secondary tympanic membrane, the most inferior part of which is cut obliquely as it bulges into the scala tympani at the region of the end of the cochlear aqueduct; *p.v.*, 'pars vestibularis' of the basal turn of the ductus cochlearis; *sac.*, sacculus; *s.e.*, saccus endolymphaticus; *s.t.*, scala tympani; *s.tr.*, transverse venous dural sinus; *s.v.*, scala vestibuli; *utr.*, utriculus; *v.*, vestibule. The arabic numerals indicate the half-turns of the cochlear duct.

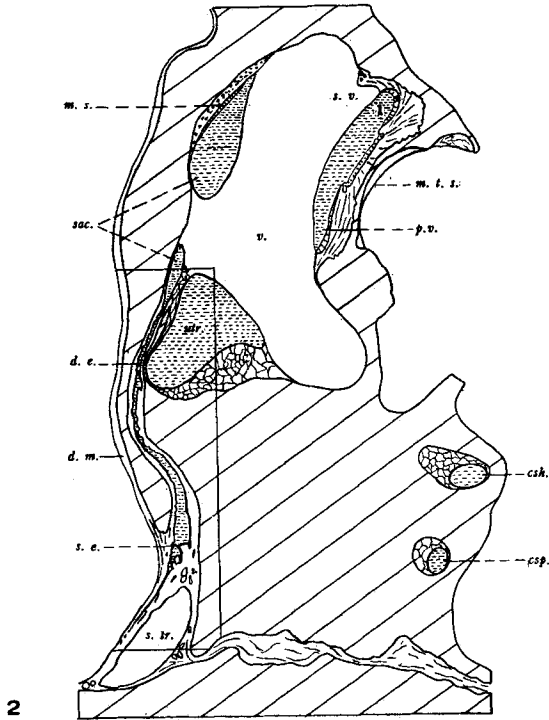
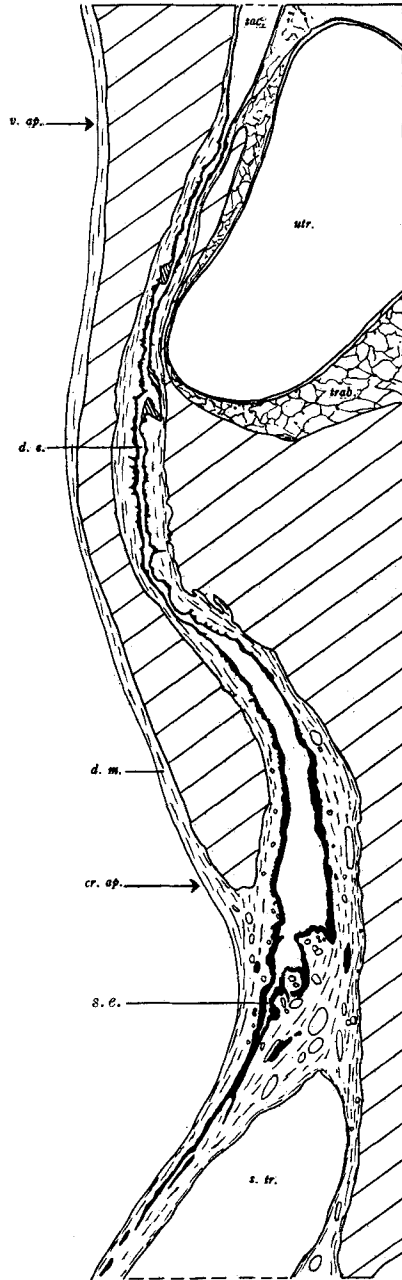


Fig. 2 Outline drawing of a single section of the internal ear of a guinea-pig. Guinea-pig 75, left ear, slide 15, row 2, section 8. $\times 15$. As in figure 1, the entire length of the lumen of the endolymphatic duct is included in the single section, and the general orientation is also similar; but there are some differences in details of the relations to other parts of the labyrinth which are described in the text matter. The part blocked out with lines is shown in figure 3 at a higher magnification. *m.s.*, macula sacculi; *m.t.s.*, secondary tympanic membrane across end of scala tympani; other abbreviations as in figure 1.

Fig. 3 Outline drawing at a magnification of fifty diameters of the part indicated in figure 2. This was drawn with an Edinger projection apparatus at 100 diameters, with especial care to the exact portrayal of the thickness and outline of the epithelium of the walls of ductus and saccus endolymphaticus. This epithelium is shown in solid black; the details of structure of typical areas are shown in the succeeding figures. The lumen of the part of the saccus overlying the transverse sinus was so narrow that it could not be shown at this magnification, so that in this part the epithelium of each wall is but half the thickness of the outline; this is likewise true of the epithelium of the distal parts of the crypts of the saccus. When followed in the series, all of the bits of epithelium which appear in single sections to be isolated are found to be in reality parts of true crypts. The small blood vessels are shown in outline as hollow; note their distribution about the region with thicker epithelium. *cr.ap.*, *v.ap.*, the arrows indicate, respectively, the cranial (or external) and the vestibular (or internal) apertures of the osseous aquaeductus vestibuli, bone being shown by oblique

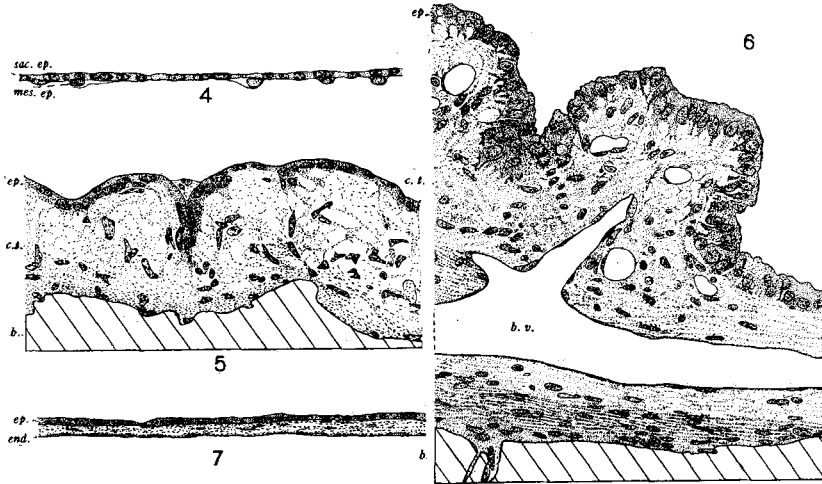


hatching as in the other figures. *d.e.*, ductus endolymphaticus; *d.m.*, dura mater; *sac.*, sacculus; *s.e.*, saccus endolymphaticus; *s.tr.*, transverse dural venous sinus; *trab.*, the trabeculated superior part of the vestibule; *utr.*, utriculus.

meter superior to that of a plane passing through both the cochlear nerve in the internal auditory meatus and the modiolar axis of the cochlea.¹ The most superior parts of the lower turns of the cochlear duct are nearly in the same plane as that of the ductus endolymphaticus, and several bits are included in one of the cases used for illustration (fig. 1), while only the junction of basal turn proper and its vestibular part show in the other one figured (fig. 2). Most of the secondary tympanic membrane is superior to the plane of the sections, as is all of the stapes. The sacculus is cut through what is almost its longest anteroposterior diameter. In the series from which figure 1 was drawn the canalis reuniens joins the inferior part of the wall of the sacculus 0.27 mm. inferior to the plane of the section sketched, and in the series from which figure 2 was made this distance is 0.72 mm. For the eleven series cut through the long axis of the ductus endolymphaticus the junction of the canalis reuniens with the sacculus averages 0.57 mm. inferior to the plane of the section including the ductus endolymphaticus, as determined by counting the intervening sections, and the range of variation is from 0.27 to 0.78 mm. The narrowed posterior portion of the sacculus which leads to the ductus endolymphaticus is so placed that its lumen is not continuous in the single section with that of the main part of the sacculus in seven of the series (as in fig. 2), but in the other four cases the condition is as in figure 1. That this difference indicates an actual variation in the relations of the parts of the membranous labyrinth with reference to the course of the ductus endolymphaticus is indicated by the fact that the four cases with a continuous lumen of the sacculus showing in the plane of section of the ductus are also the ones with the four shortest distances between this section and the entrance of the canalis reuniens into the sacculus.

¹ These have proved for me the best landmarks to orient from for sectioning, as the deeper part of the ductus itself is very difficult to locate in the block, although the relation of the vestibular end of the aquaeductus vestibuli to the crus commune makes it theoretically possible to estimate its position.

The extreme length of the combination of sacculus, ductus endolymphaticus, and saccus endolymphaticus included in the single sections through the plane of the ductus varies from 4.909 mm. to 6.204 mm., and averages 5.463 mm.; of this total length the part of the sacculus included averages 1.846 mm. For purposes of these measurements, the saccu-



Figures 4 to 7, inclusive, are drawings of typical regions of the walls in the guinea-pig of the parts indicated, and all are at the same magnification, 295 diameters, in order to facilitate direct comparison.

Fig. 4 Typical region of the lateral wall of the sacculus. This is shown for comparison because of the common statement that the epithelium of the walls of the ductus and saccus endolymphaticus is similar to that of the sacculus. *sac.ep.*, epithelium of wall of sacculus; *mes.ep.*, mesenchymal epithelial cells of the wall of the perilymphatic space of the vestibule.

Fig. 5 Typical region of the wall of the narrow part of the ductus endolymphaticus (ductus endolymphaticus proprius of the suggested terminology). *ep.*, epithelium; *c.t.*, connective tissue; *b.*, bone.

Fig. 6 Typical region of the wall of the part of the saccus endolymphaticus in which columnar epithelium is present (pars intermedia of the saccus endolymphaticus proprius of the suggested terminology). *b.v.*, longitudinally cut blood vessel; this vein empties into the transverse sinus. The lumina of this and the other blood vessels are empty because of the vascular injection of the fixation fluid. Other abbreviations as in figure 5.

Fig. 7 Typical region of the wall of the part of the saccus endolymphaticus which overlies the transverse sinus (pars distalis of the saccus endolymphaticus proprius of the suggested terminology). *ep.*, epithelium of wall of saccus; *end.*, endothelial wall of transverse sinus.

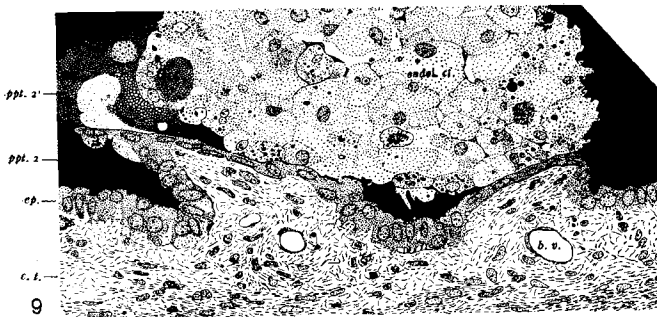
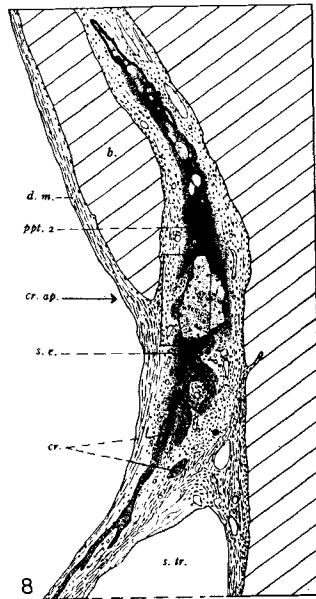


Fig. 8 Low-power drawing from a horizontal section of a guinea-pig ear, showing part of the endolymphatic duct and sac and the normal contents of the same. $\times 48$. The part marked off is shown at higher magnification in figure 9. *b.*, bone of the medial wall of the aquaeductus vestibuli, the cranial aperture of which is indicated by the arrow from *cr.ap.* *cr.*, crypts of saccus endolymphaticus; *d.m.*, dura mater; *ppt.2*, precipitate which stains very heavily with iron hematoxylin after Zenker-formalin fixation; *s.e.*, saccus endolymphaticus; *s.tr.*, transverse sinus.

Fig. 9 Detail sketch of the region marked off in figure 8, which shows the general orientation. $\times 295$. Two of the thin areas of epithelium in the region characterized by columnar epithelium are shown, and also some of the cells and precipitate which are normally present in this part of the saccus. *b.v.*, one of the small blood vessels; *c.t.*, connective tissue; *endol.cl.*, cells and cellular debris in the lumen of the saccus endolymphaticus; *ep.*, epithelium of the wall of the saccus; *ppt.2*, darkly stained precipitate in the lumen; *ppt.2'*, some of the precipitate which has bleached sufficiently in the iron-alum solution to show something of its structure.

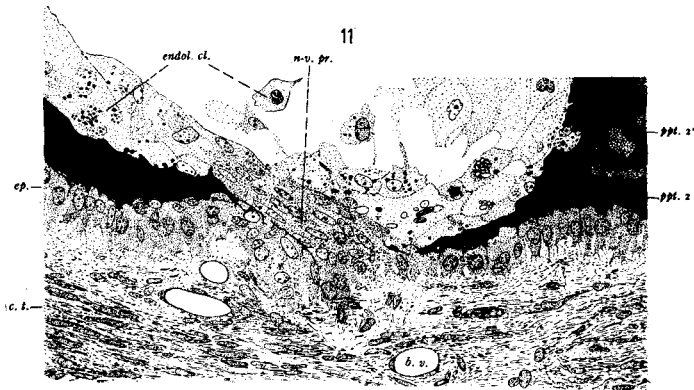
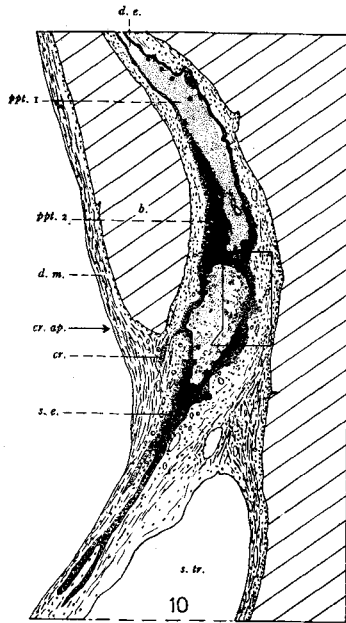


Fig. 10 Low-power drawing of a horizontal section of a guinea-pig ear, showing part of the endolymphatic duct and sac and the normal contents of the same. $\times 48$. The part marked off is shown at higher magnification in figure 11. *b.*, bone of the medial wall of the aquaeductus vestibuli, the cranial aperture of which is indicated by *cr.ap.*; *cr.*, part of a crypt of the saccus; *d.e.*, ductus endolymphaticus (the leader ends at the region of junction of the parts for which the terms ductus endolymphaticus proprius and pars proximalis of the saccus endolymphaticus proprius are suggested); *d.m.*, dura mater; *ppt.1*, loosely organized light-staining precipitate; *ppt.2*, dense precipitate which stains very heavily with iron hematoxylin; *s.e.*, saccus endolymphaticus; *s.tr.*, transverse sinus.

Fig. 11 Detail sketch of the region marked off in figure 10, which shows the general orientation. $\times 295$. The drawing shows a non-vascularized epithelial process (*n.v.pr.*) cut through its long axis, as well as typical wall and contents of the columnar epithelial region of the saccus. Other abbreviations as in figure 9.

lus was regarded as extending to the internal aperture (labyrinthine end) of the bony aqueduct, which in my material averages about 0.3 mm. distal to the entrance of the connection from the utriculus (utriculo-saccular duct). The reasons for this will become apparent later. In table 2 are given the actual measurements from ten of the series in which both the internal aperture of the aquaeductus and the saccular end of the connection from the utriculus were included in a single section so that direct measurements could be made. Only part of these are from other sections in the series in which another single section included the whole length of the ductus endolymphaticus and the rest are from other series which chanced to strike the desired features.

The resemblance to a single sac constricted in the middle part, as emphasized by Portman ('19), is noticeable in figure 1. Since the long axis of the saccus endolymphaticus is not in the plane of the sections through the ductus, the dimensions of it in these sections measured do not represent its greatest total length, so that a comparison between the ductus and saccus cannot be made from this material without resort to reconstruction. Study of the sections makes it evident that the long axis is inclined superiorly at its distal end. While the greatest vertical (superior-inferior) diameter also cannot be determined without reconstructions, it is possible by calculations from the number of sections involved to tell the greatest dimension in this direction as it would be projected on a plane at right angles to the plane of the sections. Such a dimension as determined for ten endolymphatic sacs is given in table 3. The range is rather extensive, being from 1.56 mm. to 2.66 mm., with an average of 2.23 mm. This is somewhat greater than the figures of Portman ('19), who says it is from 1.5 to 2 mm. in the vertical direction. It cannot be told from his account how this dimension was determined. In tabulating the measurements, the extent of the saccus above and below the plane of the section through the center of the length of the ductus endolymphaticus has been indicated for each one. This shows that in all cases the

sacculus extends farther superior to this plane than inferior to it. There is considerable variation with respect to this point; in one case only 9.8 per cent of this vertical extent

TABLE 1

Dimensions of some parts of the labyrinth as seen in single sections which are so oriented as to include the whole length of the lumen of the ductus endolymphaticus. Direct measurements with screw micrometer ocular (text, p. 11, and figs. 1, 2, 3)

GUINEA-PIG NO.	WEIGHT	TOTAL LENGTH OF SACCU- LUS + DUCTUS AND SACCU- S ENDOLYMPHATICUS INCLUDED IN SECTION ¹	EXTENT OF SACCU- LUS IN THE SECTION. ² LUMEN AS IN FIGURE 1; OTHERS AS IN FIGURE 2	LENGTH OF NARROW PART OF MEMBER, LABYRINTH, ³ 'DUCTUS ENDOL. PROP.' ^{4, 5}	LENGTH OF MEMBER, LABYR. FROM END OF NARROW PART TO APERT. EXT. AQ. VESTIB. = 'SUBOSTEAL' PART OF 'SACCUS' ⁵	LENGTH OF OSSEOUS AQUAEDUCTUS VESTIB- ULI = LENGTH OF DUCTUS ENDOL. OF USUAL DESCRIPTION ³	LENGTH IN SECTION OF 'INTRADURAL' PART OF MEMBER, LABYR. = SACCU- S OF USUAL DESCRIPTION ⁴	LENGTH IN SECTION OF 'SUBOSTEAL' + 'INTRA- DURAL' PARTS = SACCU- S ENDOLYMPHATICUS OF GUINEA-PIG	PER CENT OF LENGTH OF OSSEOUS AQUAED. VESTIB. THROUGH WHICH NARROW PART OF MEMBER, LABYR. EXTENDS
	<i>grams</i>	<i>mm.</i>	<i>mm.</i>	<i>mm.</i>	<i>mm.</i>	<i>mm.</i>	<i>mm.</i>	<i>mm.</i>	<i>Per cent</i>
75 Lft	175	5.573	2.182	1.388	0.785	2.173	1.218	2.003	63.9
A18 Lft	230	5.429	1.954	1.758	1.272	3.030	0.445	1.717	58.0
A25 Lft	265	5.580	1.962	1.663	1.550	3.213	0.405	1.955	51.8
A19 Lft	270	5.580	1.707	1.566	1.326	2.892	0.981	2.307	54.1
A24 Lft	335	5.003	1.660	1.382	1.342	2.724	0.619	1.961	50.7
A4 Lft	350	5.658	*2.218	1.450	0.879	2.329	1.111	1.990	62.3
A22 Lft	355	5.638	*1.388	1.313	1.289	2.602	1.648	2.937	50.5
A17 Lft	405	4.917	1.867	1.319	1.635	2.954	0.096	1.731	44.6
A16 Lft	410	4.909	*1.607	1.414	1.228	2.642	0.660	1.888	53.6
A6 Lft	440	5.649	1.923	1.547	1.298	2.845	0.881	2.179	54.4
A3 Lft	450	6.204	*1.838	1.576	1.674	3.250	1.116	2.790	48.5
Averages		5.467	1.846	1.489	1.298	2.787	0.835	2.133	53.8

¹ The figures in this column are the sums of the dimensions for each constituent part, measured as far as possible along the axis of each part, and they are therefore somewhat greater than the measurement would be if determined as the straight-line distance between these extremities of the continuous cavity (fig. 1). This is the greatest length for the ductus, but not for the sacculus and the intradural part of the sacculus. See also footnote 3.

² See text, page 17, for a statement of variations in sacculus orientation; and pages 20 and 42 for the question of place of junction of sacculus and ductus endolymphaticus.

³ Determined as sum of lengths of the two preceding parts in tabulation; this is a correct procedure because of the effect otherwise of the change in direction of the axis of the aquaeductus.

⁴ The posterior end of the long axis of this part is much superior to the plane of this section, so that this value is much less than its greatest length.

⁵ See text, page 44, for the discussion of the basis of this suggested terminology for the guinea-pig.

was below the reference plane, while at the other extreme is one with 41.7 per cent below. The average for the ten cases of the percentage of the vertical extent superior and inferior to the plane of the section through the ductus endolymphaticus is 73.6 per cent and 26.4 per cent, respectively. In other words, the saccus endolymphaticus on the average extends approximately three times as far above this plane of the ductus as it does below it. This differs from Portman's diagram, which shows it as extending equally above and below.

TABLE 2

Direct measurements in single sections of the distance from the internal (vestibular) aperture of the bony aquaeductus vestibuli to the nearest angle of the junction of the utriculo-sacculus duct with the sacculus (text, p. 20 and p. 42)

GUINEA-PIG NO.	WEIGHT	DISTANCE	GUINEA-PIG NO.	WEIGHT	DISTANCE
	<i>grams</i>	<i>mm.</i>		<i>grams</i>	<i>mm.</i>
75 Lft	175	0.265	A17 Lft	405	0.261
A18 Lft	230	0.330	A16 Lft	410	0.271
A19 Lft	270	0.267	A6 Lft	440	0.397
A20 Lft	275	0.216	A3 Lft	450	0.425
A24 Lft	335	0.347			
A5 Lft	390	0.257			
			Average of ten.....0.304		

At its vestibular end the ductus endolymphaticus is separated by a very thin lamina of bone (lateral wall of the bony aquaeductus vestibuli) from the body of the utriculus in the region where it is joined by the superior sinus and by the non-ampullated end of the horizontal semicircular canal. It is somewhat below the inferior margin of the fossa subarcuata (floccular fossa). The relations of the ductus and saccus to the immediately adjacent structures, including the transverse sinus and the dura, will be described later in this account.

The part blocked off in figure 2 was drawn in outline at a magnification of 100 diameters, with especial care as to the thickness of the epithelium and the size and shape of the lumen as seen in the section. This is reproduced at fifty

diameters as figure 3. No attempt was made to show histologic detail in this sketch, but simply location and proportions. The content of the lumen is omitted for the sake of clearness of outline. Details of both structure and content will be shown in other figures.

The lumen of the membranous part, as may be distinctly seen in figure 3 and on a smaller scale also in figure 1, has a more or less uniform narrow diameter from the internal

TABLE 3

Extent of the saccus endolymphaticus inferior to and superior to the plane of the approximately horizontal section passing through the whole length of the lumen of the ductus endolymphaticus. The figures are based on the number of serial sections involved (text, p. 20)

GUINEA-PIG NO.	WEIGHT	EXTENT INFERIOR TO PLANE OF DUCTUS	EXTENT SUPERIOR TO PLANE OF DUCTUS	TOTAL OF SUPERIOR-INFERIOR EXTENT	PER CENT INFERIOR TO PLANE OF DUCTUS	PER CENT SUPERIOR TO PLANE OF DUCTUS
	<i>grams</i>	<i>mm.</i>	<i>mm.</i>	<i>mm.</i>	<i>Per cent</i>	<i>Per cent</i>
75 Lft	175	0.940	1.530	2.470	38.1	61.9
A18 Lft	230	0.220	2.020	2.240	9.8	90.2
A25 Lft	265	0.360	1.200	1.560	23.1	76.9
A19 Lft	270	0.720	1.520	2.240	32.1	67.9
A24 Lft	335	0.460	2.020	2.480	18.5	81.5
A4 Lft	350	0.660	2.000	2.660	24.8	75.2
A22 Lft	355	0.675	0.945	1.620	41.7	58.3
A17 Lft	405	0.300	2.000	2.300	13.0	87.0
A16 Lft	410	0.620	1.540	2.160	28.7	71.3
A3 Lft	450	0.875	1.700	2.575	34.0	66.0
Average values		0.583	1.647	2.230	26.4	73.6

(vestibular) aperture of the osseous aquaeductus vestibuli to somewhat beyond the midpoint of the same, beyond which it expands more or less abruptly (figs. 1 and 3 illustrate fairly well the variations in the rate of this expansion) into a dilated portion lying within the cavity of the distal part of the bony canal and is continuous, without marked change in size in the plane of section, with the lumen of the part lying between the layers of the dura near the external (cranial) aperture of the aqueduct. The lumen of the portion overlying

the transverse sinus, however, is usually quite narrow, and for much of its extent frequently is merely a potential space with the lateral and medial walls in close apposition, as is the case in the preparation from which figures 2 and 3 were drawn. The lumen of this part was too narrow in this case to be shown distinctly in the sketch. In figure 1 can be seen a distinct lumen in this part, and in some of the other animals it is even wider. The collapse to a potential space of the lumen of this distal part as seen in some of my series may be due to osmosis and diffusion during the preparation of the material, since none of the surrounding parts are so attached as to support its walls in this direction, but it seems quite probable that it is never as wide normally, even in life, as the lumen of the part near the aperture of the aquaeductus.

The length of the narrow part of the lumen of this membranous part in the eleven favorably cut preparations varies from 1.313 to 1.758 mm., with an average length of 1.493 mm. (approximately 1.5 mm.). The length of the bony aquaeductus vestibuli in these same cases varies from 2.173 to 3.250 mm., and averages 2.782 mm. (approximately 2.75 mm.). The percentage of the length of the bony canal through which the narrow part of the membranous ductus extends varies from 44.6 per cent to 63.9 per cent, with an average of 53.8 per cent. In only two of the eleven cases was the narrow membranous part less than half as long as the bony canal, and the average is the basis for the statement already made above, that the narrow part usually extends somewhat beyond the midpoint of the aquaeductus. The measurements on which this and other summaries in the text matter are based are given in detail in table 1.

The relative thicknesses of the epithelial wall of the various parts of the ductus and saccus endolymphaticus as shown in figure 3 are typical of these parts in the guinea-pig. Figures 4, 5, 6, and 7 are reproductions at a magnification of 295 diameters of careful drawings at a magnification of 885 diameters of typical regions of the epithelium and the subjacent tissue of the several parts shown in figure 3, although

not from the same section that this outline sketch was made from. A glance at these figures is sufficient to show that there are essential histologic differences in the several regions, and that these differences are much greater than has hitherto been reported for any animal. For this reason, a detailed description of each part is required. Measurements were made of the thickness of the epithelium in at least ten places for each region; the places measured were selected with especial regard to the plane of section with reference to the epithelium at the particular part measured in an effort to have comparable thicknesses by always measuring at right angles to the surface. The diameters in the plane of section parallel to and vertical to the surface of the epithelium were determined for at least ten typical nuclei in each region, and care was taken in each case to be certain that the section contained the largest parts of the nucleus. The averages and the extreme ranges of the epithelial thicknesses and the average nuclear dimensions are summarized in table 4, and are utilized below in the descriptions of each region.

A typical region of the wall of the sacculus is shown (fig. 4) for comparison only, since so many of the current descriptions speak of the epithelium of the ductus and saccus endolymphaticus as similar to that of the sacculus and utriculus. As is very well known and as may be seen in figure 4, this epithelium of the sacculus is simple squamous in type; in my material ten vertically cut regions selected from various parts of the wall ranged in thickness from 1.7 to 4μ and averaged 2.6μ . Ten nuclei averaged 2.1 by 7.3μ in the plane of section, the long axes being of course always parallel to the surface. A part of the 'free' wall of the sacculus was sketched as representative, because this forms the greater part of its total extent exclusive of the macular area; if an 'attached' area away from the macular region had been sketched, a quite thin layer of areolar tissue would be seen between the epithelial wall and the bone, but not by any means comparable in thickness to that adjacent to the ductus endolymphaticus in even the narrowest part of the aquae-

ductus vestibuli. The few cells that are seen in figure 4 'beneath' the saccular epithelium are mesenchymal epithelial cells of the wall of the perilymphatic space.

A typical region of the wall of the narrow part of the ductus endolymphaticus is shown in figure 5. The walls of the region of expansion just beyond the narrow part are also of the same structure. The surface of this part is more or less irregular and is occasionally sharply depressed, due to the presence of simple crypts, some of which are as much as 40 μ deep. One such crypt is included in the region sketched.

TABLE 4

Thickness of the epithelia and average diameters in plane of section of the nuclei in each region. Averages based on ten or more measurements in each region, with especial care to avoid obliquely cut epithelium (text, p. 25)

	THICKNESS OF EPITHELIUM			AVERAGE OF DIAMETERS OF NUCLEI	
	Average	Minimum	Maximum	Parallel to surface of epithelium	Vertical to surface of epithelium
	μ	μ	μ	μ	μ
Sacculus (away from macular area).....	2.6	1.7	4.0	7.3	2.1
Narrow part of ductus endolymphaticus....	3.7	2.2	4.8	7.5	3.0
Region of columnar epithelium.....	18.6	14.4	23.6	4.5	7.3
Distal part of saccus endolymphaticus.....	4.4	3.5	5.6	7.5	2.8
Thin areas in the columnar epithelial region	3.8	2.2	¹	9.0	3.0

¹ Measurements for such areas are based on distinctly thin parts only; for the nature of transition zone to columnar epithelium, see text, p. 30.

Its lumen was so cut that its outline is apparent upon focusing for the upper surface of the section, while the surface of its wall is included in the deeper part of this 10 μ section. A tangentially cut piece from the outer surface of another crypt is also seen near by. The presence of these small crypts and the wavy surface give the epithelium of the wall of this part, at first inspection of the histologic preparations, the appearance of greater variation in thickness than appears to be actually the case upon careful comparison of serial sections when the differences in obliquity of the plane of section with reference to the surface of the wall in closely ad-

jacent areas can be taken into consideration. The region selected for the portrayal of this wall (fig. 5) was chosen with the view of including some of the extremes in appearance; the thinner portions of the part shown are the ones that best portray the real structure of the type of epithelium present. The connective tissue underlying this epithelium is of a very loose areolar type with very few blood vessels, and in most of the region it is impossible to distinguish in my preparations anything which can be termed a basement membrane, although occasionally for short distances a poorly defined layer resembling a basement membrane can be seen. Processes extend into the loose areolar tissue for variable distances from the deep surface of many of the epithelial cells of this region; some of these processes are as much as 6 to 9 μ in length. Measurements of this epithelium in ten places in which the factor of obliquity was ruled out as far as possible gave an average thickness of 3.7 μ , with a range of from 2.2 to 4.8 μ . The diameters in the plane of section of ten nuclei averaged 3 by 7.5 μ , with the long axis in every case parallel to the surface.

From the above description, it is apparent that this epithelium of the narrow part of the ductus endolymphaticus may best be classified as simple squamous in type, and in this respect it may be compared to that of the sacculus, from which; however, it differs essentially in being distinctly thicker and in having both basal processes and simple crypts which are in marked contrast to the uniformly regular deep surface of the saccular epithelium. The very rarely observed small evaginations of the walls of sacculus and utriculus are quite different in character from these true epithelial crypts. The region of transition between these two epithelia which are so essentially different in histologic structure, although both of a simple squamous type, occurs constantly very close to the internal (vestibular) aperture of the aquaeductus vestibuli, which is, in the guinea-pig, as mentioned previously in the description of the relations, about 0.3 mm. distal to the junction of the duct from the utriculus. This

point will be considered further in the discussion of my observations.

The dilated part of the membranous labyrinth which is located in the distal part of the bony aqueduct and between the layers of the dura mater near its external aperture has the greatest part of its wall surfaced with a distinctly columnar type of epithelium, which is continuous with that of the squamous type just described by way of a zone of epithelium of gradually increasing thickness on the wall of the larger end of the region of expansion. This transition in thickness may be seen in figures 3, 8, and 10, but no part of this region has been portrayed in detail, since it is distinctly intermediate in character between two areas for which careful descriptions and drawings are given. The wall of the dilated part which may be said to be characterized by the simple columnar epithelium is complicated by the presence of small areas of squamous epithelium, by small non-vascularized projections into the lumen, and by larger vascularized processes into the lumen and crypts into the surrounding tissue. Figures 6, 8, 9, 10, and 11, besides the outline sketches already considered, portray the various features of this region. A typical area of the wall of this part is shown in figure 6, while the other figures include both typical and certain of the peculiar features of the region. The orientation of the areas shown in detail in figures 9 and 11 is apparent upon inspection of the lower-power sketches in figures 8 and 10, respectively, and figure 6 shows a part of the lateral wall of the saccus just beyond the limits of the bony canal. The contents of the lumen, similar in character to those shown in the other figures, have not been included in figure 6, although present, in order to present a drawing of the wall itself more directly comparable with those of the other parts portrayed in figures 4, 5, and 7. The irregular arrangement of the epithelium at the bottom of both depressions shown in figure 6, which might be interpreted from a single section as indicating a stratified or at least a pseudostratified character, is in reality due to the fact, which serial sections

reveal, that the edges of the proximal ends of large epithelial crypts like some of those seen in low power in figures 3 and 8 are included. The walls of these crypts, many of which are quite elongated and even branched, when favorably cut for observation, are always lined by the same type of columnar epithelium as shown for the major part of the wall in this region.

This columnar epithelium has distinct basal processes which for the most part extend varying distances into the underlying connective tissue without any appearance of terminating on any condensation which may be termed a basement membrane, but there are occasional small areas where the processes and even the deeper ends of whole cells do rest upon what one may call a very indistinct basement membrane. Both conditions are present in the regions sketched in figures 6 and 9. For the most part, cell boundaries are very indistinct except occasionally where some of the material in the lumen, to be described later, appears to have extended into crevices between the superficial ends of cells and has retained in its characteristic way the hematoxylin stain during the bleaching with iron-alum solution. The nuclei are not at a uniform level in the cells—some are near the free surface, others near the deep or basal part, and many are near the center of the cells. Most of the nuclei are roughly oval in outline, but some approach a circular outline even in cells that appear to be cut in the long axis. It is quite common for a nucleus to have three or four nucleoli. Measurements of the thickness of ten vertically cut areas of this columnar epithelium of the dilated part, including the basal processes, ranged from 14.4 to 23.6 μ , with an average of 18.6 μ , and the averages for ten nuclei of the diameters in the plane of section were 7.3 by 4.5 μ , the long axes being vertical to the surface of the epithelium.

The location of two small areas of squamous epithelium, mentioned above as occurring in this dilated part, is shown in figure 8 and the details of the region in figure 9. The thinnest place observed was only 2.2 μ thick, but the average

thickness of ten places was 3.8μ and the nuclei averaged 3 by 9μ in the plane of section, with the long axes parallel to the surface. The two areas seen in figure 9 appear to be separated by an 'island' of columnar epithelium, but it is apparent upon following these through the adjacent sections of the series that there are really two thin areas surrounded by the typical columnar epithelium.

Toward the left in figure 9 the epithelium of the thin area may be seen to be continuous with that of a projection into the lumen at the region of junction with the columnar epithelium. An indication of the same type of formation is evident toward the right end of the same figure for the other thin area, and the adjacent sections show this to be actually the case. A much larger, but in most respects essentially similar, epithelial process is shown in figures 10 and 11; however, it is in this case not associated with an adjacent thin area of epithelium. Careful inspection of figure 11 is necessary to distinguish the outline of this epithelial process from the adherent free cellular content of the lumen; and at the region of the apex of this process, which is toward the left, it was even difficult when directly observing the specimen itself to determine the exact demarcation of the process. The possibility that such processes may be the source of part of the free cells of the lumen will be considered after the contents of the lumen have been described. In figure 11 the demarcation between the epithelial surface of the wall proper and the adjacent overhanging surface of the epithelial process is clearly apparent, due to the presence of a small amount of the dark-staining precipitate in the lumen. This process is cut through its central part, and the section was selected for sketching because of this fact. The appearance is not due to a tangential cut through the epithelium of a larger process with a single layer of surface cells, but the structure is a true non-vascularized papilla-like process formed by a heaping up of more or less elongated and somewhat flattened cells. Inspection of the nuclei of these cells, as seen in figure 11, shows them to be distinctly different in

shape and size from those of the adjacent columnar epithelium. In two of the series on hand, both being unfortunately among those prepared by the method for locating the Prussian-blue granules and accordingly not well suited for the final interpretation of histologic detail, there are present what appear to be trabeculae of epithelium only extending across the lumen of this part of the endolymphatic sac. Upon the basis of such material I hesitate to make a very positive statement, but these trabeculae appear to be essentially like the non-vascularized epithelial papillae just described, except that they are attached to each other in such a manner as to form trabeculae.

The size and location of several of the large crypts associated with the wall of this dilated region are shown in figures 3, 8, and 10. Some of these are sectioned more or less lengthwise and include the region of junction with the main part of the wall, while others chance to be so oriented that the region of attachment is not included in the section, so that they appear as isolated parts. In all cases, however, such bits when followed through the series are found to be continuous with the wall proper. That these are true crypts, and not simply due to folding or wrinkling of the wall of the region, is readily determined by following them through the serial sections. Reconstruction is, of course, necessary for the determination of all dimensions of these crypts, and such reconstruction has not been done. These crypts vary considerably in length, but extend quite uniformly in the same general direction, the tip or free end of each being distalward, with reference to the main part of the membranous labyrinth, to the base or attached end. With reference to the animal as a whole, this means that the crypts extend caudalward from their region of origin. The epithelium which forms the walls of these crypts is very similar to the columnar epithelium typical of the wall of the main part in the region of attachment, which has been fully described already. The lumen of a crypt typically tapers until it is very small, and frequently toward the distal end may be described as potential only.

Between the attachments of the crypts just described the wall frequently projects as a whole into the lumen, although the apparent height of most of these processes is due in large part to the fact of the extensions of the lumen into the adjacent crypts. Such processes may be seen in figures 3, 6, and 8. The epithelium covering them is typical of the region.

A well-vascularized areolar connective tissue surrounds this specialized columnar epithelial part. The fibrous content of this areolar tissue is distinctly greater than that of the tissue surrounding the narrow part of the ductus endolymphaticus, but near the epithelium it is what may be termed a very loose areolar tissue. The endosteal layer of the dura mater is continuous with a dense fibrous tissue next to the bone of the cranial end of the aquaeductus vestibuli; this constitutes the endosteum of the bony canal and in the narrowed part is a very thin layer. This dense areolar tissue is separated from the epithelial wall at all parts by the looser tissue, which also is interposed between the medial wall of that part of the columnar epithelial area which extends beyond the cranial aperture of the aquaeductus and the firm tissue of the meningeal layer of the dura mater. This is what Sterzi ('09) has termed the perisaccular tissue.

As may be seen in several of the figures, there are many small blood vessels in the loose areolar tissue adjacent to the parts of the columnar epithelial area. Only a few vessels are in direct contact with the epithelial cells; for the most part, even those of capillary size are separated from the basal surface of the epithelium by a small amount of quite loose fibrous tissue. Special vascular injections for the study of the connections of these vessels to the others of the region have not been made, as stated in the review of the literature, so that very little can be stated now concerning this point. In figure 6 one of the larger veins is included in longitudinal section, and when followed in the series it can be seen to open into the transverse sinus. This is in agreement with Sterzi's observation in the human, and Portman's in the guinea-pig.

The wall of that portion of the saccus endolymphaticus which overlies the transverse sinus is for the most part composed of epithelium which is of a rather thick simple squamous type and corresponds quite well with the usual descriptions of saccus endolymphaticus epithelium. In places it may be termed a low cubic epithelium. Measurements of the thickness of this epithelium in ten regions selected as vertically cut ranged from 3.5 to 5.6 μ , with an average thickness of 4.4 μ ; the nuclei average, in the plane of section, 2.8 by 7.5 μ . A section from the lateral wall of this part, including the adjacent wall of the transverse sinus, is shown in figure 7. That this epithelium is essentially different from that of the other regions, shown in figures 5 and 6, is very evident. The junction of this type of epithelium with the columnar type described above is a somewhat gradual transition and usually occurs at or near the anterior margin of the transverse sinus. The usual relations of the external (cranial) aperture of the aquaeductus vestibuli and the margin of the transverse sinus are as shown in figures 8 and 10; but in some cases the anterior edge of the sinus extends forward until it is in the medial wall of the distal end of the bony canal, and in these circumstances the transition between columnar and squamous epithelia may be located over the transverse sinus rather than at its margin. Inferiorly, this boundary between the parts is nearer the cranial aperture of the aquaeductus than it is superiorly; this is apparently associated with the closer approach of the sinus to the slit-like aperture inferiorly. While the walls of this distal part of the saccus are usually quite smooth and regular, being much like the customary descriptions of the whole saccus, there are occasionally individuals in which true epithelial crypts into the surrounding tissue are present, as well as some folding of the wall which may give a similar appearance in single sections.

Frequently, the saccus endolymphaticus extends completely across the transverse sinus with its most distal part posterior to the sinus; in a few of these cases the epithelium of this distal tip is quite like that of the columnar area, and a few short crypts may be present.

The connective tissue adjacent to the walls of the squamous epithelial area overlying the transverse sinus is of a relatively firm nature and rather scanty in amount as compared to that of the region near the orifice of the aqueduct. It blends medially with the firm tissue of the meningeal layer of the dura mater and laterally with the tissue of the wall of the sinus. It is also relatively poorly vascularized as compared with the region near the orifice of the aqueduct, although a few small vessels may be found near the epithelial wall of this part of the saccus. In those cases mentioned above, where the saccus projects posterior to the transverse sinus, the connective tissue adjacent to this distal extremity is looser in character and contains more small vessels.

CONTENTS OF LUMEN

Numerous free cells and much precipitate are present in sections of the normal guinea-pig endolymphatic duct and sac. This is in very marked contrast to the lumina of all the other parts of the membranous labyrinth, where free cells of any kind are but rarely observed and where the precipitate formed by the endolymph upon fixation is very scanty in amount. In the endolymphatic duct and sac the cells and the precipitate are not uniformly distributed, but are almost entirely limited to the lumen of that part which I have described as characterized by having a columnar epithelial wall and of the proximally adjacent region of the expanding lumen. While in the usual terminology this must be termed the distal part of the ductus and the proximal part of the saccus, in the terminology which is to be proposed on the basis of this work (see below) these free cells and the precipitate may be described as being in the proximal and inter-

Fig. 12 A few of the cells found in the lumen in sections of the saccus endolymphaticus (and dilated part of the ductus of the usual terminology) of normal guinea-pigs. The original drawings were made with the aid of an oil-immersion lens at 1400 diameters, and are reproduced as half-tone illustrations at a magnification of 1260 diameters. For the description and discussion of these cells, see the text matter, pages 34 to 40.

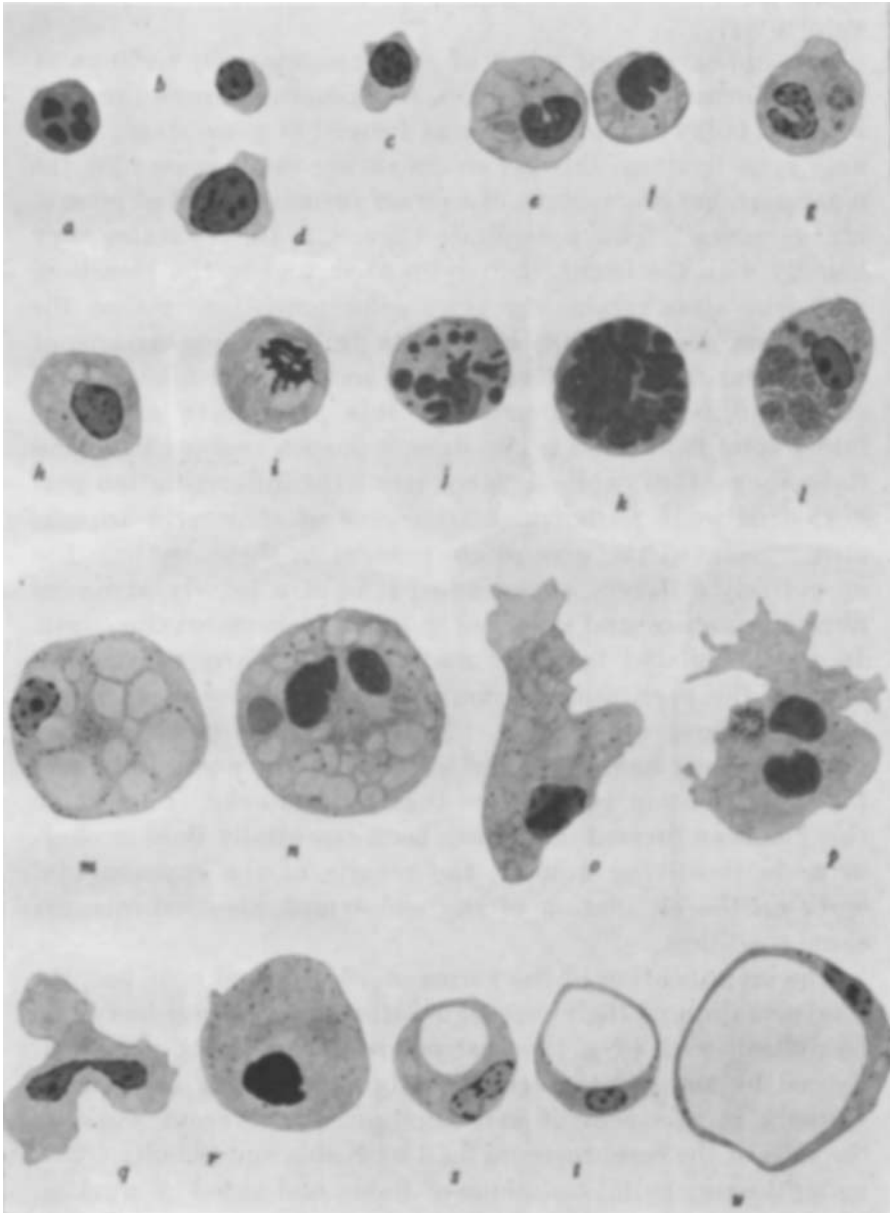


Figure 12

mediate parts of the saccus endolymphaticus proprius of the guinea-pig.

The appearance of much of the precipitate in sections of Zenker-formalin material gives the general impression that a rather bulky colloidal 'gel' was formed at some stage, probably upon fixation; this gel shows rather small spaces for the most part, but has regions of coarser formation and of irregular 'vacuoles.' This precipitate (figs. 8, 9, 10, 11) stains very heavily with the iron-hematoxylin used, and in the bleaching with iron alum retains the black color much longer than the cytoplasm does, and in some parts fully as long as any of the nuclear material. Even in $10\ \mu$ sections which have good cellular differentiation parts of this precipitate are absolutely solid black, and in the drawings such regions have had to be shown thus, while in other parts the differentiation permitted of some portrayal of the general structural appearance. Some of the precipitate present in these sections has an entirely different appearance; it is of a loosely arranged fibrinous nature and does not retain the hematoxylin stain. In the acidulated formalin material with carmine staining none of the precipitate is distinctly organized and all has a more fibrinous appearance. Many cells and much cellular débris are surrounded by, and included in, the precipitate, and their distribution is evidence that the material from which this has been formed must have been essentially fluid in character in the living animal; the results of the experimental work on the circulation of the endolymph also indicate the same condition.

The identification of the forms of white blood cells and the interpretation of their genetic relationships is well known to be difficult with even the best of special methods, as is evidenced by the great diversity of opinion among the special workers in this field of investigation. The recent work on the cells of the cerebrospinal fluid by Kubie and Schultz ('25), using the supravitral technique of Sabin and aided by working in the laboratory where first-hand training in the methods of Sabin and Cunningham and their co-workers was possible,

has emphasized the unreliability of interpretation of such cells by their appearance in fixed preparations alone. Accordingly, in view of the fact that I have available as yet only fixed material in sections prepared with general stains only, I feel that any extensive discussion of the cellular elements in the saccus endolymphaticus would be of little value. Therefore at this time only a series of sketches of a few of the cells present in the sections and a very brief discussion will be attempted. It is, I believe, feasible to obtain these cells in condition to make a supravital study of them, and this should make possible eventually a real classification.

In figure 12 are shown half-tone illustrations of careful crayon drawings of some twenty-two of these cells made at a magnification of 1400 diameters, using an oil-immersion lens. They are reproduced at nine-tenths original magnification. While these are not all from the same endolymphatic sac, they are all from normal guinea-pigs and, with the exception of one cell, all are in material prepared by the same technique (method 1). These sketches do not show the relations of the cells to the walls and to other cells as do figures 9 and 11; in fact, an effort was made to find cells which were not in contact with others so as to portray the shape when free. In sectioning parts of cells have, of course, been cut away, and in selecting ones to sketch only those were chosen which happened to be favorably oriented in the section for displaying the essential parts. Disintegrating cells and cell fragments have not been sketched, although plentiful in the material. In figures 9 and 11 the cells and cellular débris and the precipitates which chanced to be in the lumen of the parts sketched have been shown in actual relation to each other; in comparing the cells of these figures with those in figure 12, it is necessary to keep in mind both the difference in magnification and in method of representation; one is pen-and-ink work reproduced by a line-engraving process, the other is half-tone reproduction of crayon work.

Some of these cells are unquestionably white blood cells, and some of the cells are definitely phagocytic in nature, but

concerning the source of these and their proper classification I am very uncertain, as indicated above. Of the cells shown in figure 12, *a* is without doubt a polynuclear leucocyte, and *b*, *c*, and *d* look very much like small, medium, and large lymphocytes, respectively. Cells *e*, *f*, and *g* have nuclei of the so-called 'transitional cell' shape; the cytoplasm in all is more or less vacuolated and all contain 'cell inclusions.' In cell *g* the clusters of dots shown in the cytoplasm represent brown particles, probably pigment granules. These cells may belong to the monocyte group of Sabin and her pupils. Cells *h* and *i* may also belong to this group, as their size is within the correct range. Cell *i* was sketched not as typical of many seen, but in order to place on record a cell observed in a stage of mitotic division in the lumen of the saccus endolymphaticus. In this cell the diaster phase of mitosis is present; the axis of the spindle is so obliquely placed with reference to the plane of the section that the portion of the other group of chromosomes which was not removed by the knife cannot be shown without overlapping in the sketch, but can be readily seen in the preparation itself by proper focusing. This cell is proof that at least not all of the cells present are in a condition of degenerative changes.

Many cells of the general character of *j* and *k* have been observed; these cells are usually regular in contour and contain various numbers of very dark-staining masses of material which may, as in *k*, completely obscure all the other cell features. A nucleus can be seen in *j*, partially obscured by some of the dark-staining material. Of the substance of which this relatively massive cellular content is composed I do not have any data other than its appearance; it seems possible that it is some of the same material which when precipitated in the lumen of the saccus stains so darkly with the iron hematoxylin. It has probably been phagocytized by these cells, whatever its source, although it is, of course, possible that it is a product of the cellular activity. In some animals there are many cells with large numbers of ingested brown particles, probably pigment granules. One of these

is figured as *l* of figure 12; it was prepared by the second method, which gives a better view of pigment granules. A few small vacuoles are sometimes present and, except for the character of the inclusions, these cells resemble in appearance cells *j* and *k*.

Cells of the general appearances of those shown as *m* and *n* and as *o* and *p* are quite numerous, and it seems probable from the intergradations that can be observed that these may all represent various phases of the same type of cell. All are quite large and of about the same general size; in all the cytoplasm contains several vacuoles, which may be either small or quite large, and there are discrete granules in the cytoplasm between vacuoles. In cell *p* there is also a clump of brown granules, probably pigment. Although cells *o* and *p*, by their outline, show definite evidence of having possessed the power of ameboid movement, while *m* and *n* are rounded up, it is possible that this may only be a difference in the phase of activity when fixed rather than an essential difference. The nucleus in such cells is usually, but not always, excentric in position, as in *m* and *o*. As in *n* and *p*, two nuclear masses may be present; of the significance of this I am very uncertain, but it is possible that the condition shown in cell *q* is related to this and that segmentation of the nucleus with or without division of the cytoplasm may occur in these cells. Cell *q* seems otherwise to belong in the same category with *o* and *p*. Cells of the type represented as *r* are of frequent occurrence, and although vacuoles are not present, its size indicates that it may also belong in this group. The nuclei of some of these cells stained so intensely that no structural details could be seen in preparations in which most of the nuclei were well differentiated.

The cells figured as *s*, *t*, and *u* are representative of a group frequently observed, and I have arranged these three in what seems to be the most probable growth sequence. All are of the 'signet-ring' shape in section, and appear to have consisted of a more or less thinned-out cytoplasmic shell containing a large vacuole or globule of some substance which has

quite completely disappeared during the preparation of the material. Such a formation may, of course, have been due to a coalescence of several smaller vacuoles, like those seen in *m* and *n*, or may have been due to the continued growth of an originally small separate droplet of some substance. As in *s* and *u*, a few 'secondary' small vacuoles are frequently present in the cytoplasmic shell and a few discrete granules, which look like those in the cells *m* and *n*, are to be seen. While these may really belong in the same general group with the cells like *m*, *n*, *o*, *p*, and *q*, they are so striking in appearance that it has seemed best to describe them separately.

Some of the cells, especially those shown in figures 9 and 11, look much like disintegrating desquamated epithelial cells, and there are occasionally observed in the walls of the ductus endolymphaticus more or less swollen vacuolated cells which were possibly undergoing changes that would have led to desquamation into the endolymph, but I have seen none in which I can be certain as to the fate of such cells. Under the description of the non-vascularized epithelial processes of the wall of the saccus, attention was called to the gradual transition in appearance from cells of the process to adjacent but free cells of the lumen, and the possibility was mentioned that this might be the source of some of these free cells. On this point, also, I do not feel that the evidence is sufficient to justify any positive statement, and I can simply call attention to the possibility. In concluding this section it seems proper to indicate again that the employment of the best of the special hematologic methods is necessary before any real classification can be made of these cells which are a normal content of the endolymph in the ductus and saccus endolymphaticus of the guinea-pig.

DISCUSSION AND CONCLUSIONS

Ordinarily, morphologic differentiation forms the primary basis for descriptive terms, and the relations to adjacent structures serve only as a basis for secondary divisions of

things morphologically similar. Differences in the size of the lumen or in the rate of change in size have usually been considered as more important than the differences in structure of the wall in the case of most of the hollow organs, but both are rated above the relations to adjacent parts. For most of the membranous labyrinth as well as for larger organs these principles of terminology have been used, and it seems proper, therefore, to apply them also to the parts of the labyrinth under discussion. An attempt to answer certain fundamental questions may serve to indicate the important facts.

First, what are the characteristics of the lumen? There is an elongated very narrow part of relatively constant size which has a quite definite infundibuliform expansion into dilated portions at either end; these junctions of the uniform narrow part and of the regions of expansion are quite definite and easily recognized and constitute fundamental characteristics. In the dilated portions the changes in size and shape of the lumen are gradual and not definitely demarcated, except of course where it extends out into the crypts.

Secondly, how many morphologically distinct zones of the wall are there and what are the limits of each with reference to the changes in the lumen? In the customary description, except for the macular area of the sacculus, the walls of the entire combination of parts under consideration have not been differentiated morphologically; whereas, as described above, four zones may be readily distinguished on the basis of structure alone (figs. 4 to 7). The details of the differences need not be repeated here. The boundary between two of these zones is located very close to or exactly at the vestibular end of the elongated narrow part. On the contrary, the distal end of the narrow part does not coincide with any structural change in the wall, and most of this region of expansion has a wall very similar to that of the narrow part, and only after the lumen is quite enlarged does the gradual transition in structure occur between parts that are very distinct morphologically (figs. 5 and 6). Finally,

the change between parts of the wall that look like those in figures 6 and 7 takes place rather gradually at about the middle of the region where the lumen has the greatest potential size, although it may be quite collapsed horizontally in the preparations.

Thirdly, what are the essential relations to adjacent parts and how are these correlated, if at all, to the variations in lumen and in structure of the walls? The walls and ends of the aquaeductus vestibuli, the dura mater and the transverse dural venous sinus are the adjacent structures of descriptive importance, aside from the intravestibular parts of the labyrinth. The vestibular or proximal end of the elongated narrow part of the lumen and the change in wall structure between parts as illustrated in figures 4 and 5 both occur just at the internal or vestibular aperture of the aquaeductus vestibuli. The junction with the duct from the utriculus, which is about 0.3 mm. from this aperture, is the only feature which can be interpreted as not indicative of this coincidence of three factors determining the real boundary between sacculus and ductus endolymphaticus. The fact that there is a distinct histologic change in the wall of the membranous labyrinth at the region of the internal aperture of the aqueduct is a new point in the dispute concerning the interpretation of this region, and constitutes an additional argument in favor of the views of Hasse and his followers that the essential relations in mammals are the same as in the other classes of vertebrates. In other words, the ductus endolymphaticus arises from the sacculus, and not from the junction of ducts from the sacculus and the utriculus, and the utriculo-saccular connection, while somewhat longer and narrower than in some forms, opens really into the sacculus near the orifice of the endolymphatic duct. The history of this question need not be considered in detail in this paper, but as indicative of the fact that it is not yet considered as settled the recent dispute between Wittmaack ('24 a, '24 b) and Alexander ('24) may be mentioned. To these papers reference may be made for a more complete discussion.

The external or cranial aperture of the aquaeductus vestibuli does not, however, coincide with either any marked change in lumen or in structure of the wall of the membranous parts. The transition between parts of the wall of the structure illustrated in figures 5 and 6 occurs well beyond the enlargement of the lumen, and at the same time a considerable portion of the wall characterized by columnar epithelium is within the osseous canal. The wall of that part of the membranous structure which is between the layers of the dura mater near the end of the aqueduct has exactly the same structure as that within the distal part of the bony canal, and the transition to a wall of the type illustrated in figure 7 appears to be more correlated with the region where the saccus begins to overlap the transverse sinus than with anything else; but, as already indicated, this correlation is not very close. And yet there seems to have been almost universal agreement that the external aperture of the aquaeductus vestibuli should be considered the boundary between the parts termed ductus and saccus endolymphaticus. The lumen of the bony aqueduct does enlarge in the guinea-pig at the same region where that of the membranous cavity enlarges, on the average slightly beyond the middle of the whole length of the osseous channel, and I suppose that one may consider that it is this place which has been meant as the boundary, but this has not been made clear or given emphasis in even the special descriptions. There is no doubt that the BNA term 'apertura externa aquaeductus vestibuli' was intended to designate the slit-like opening in the human, and not the junction of the narrow channel with the enlargement. Sterzi ('09) recognized some of the difficulties in terminology in this region in the human, and, on the basis of a description of the posterior surface of the pyramid much more complete than the usual one, he proposed the term 'recessus saccularis' for the enlargement of the cranial end of the aqueduct and that the term apertura externa be applied then to the orifice of the narrow part in the apex of this recess, which, as he observed, contains the proximal end of

the true saccus. For the slight depression in which the rest of the saccus is located he proposed the term 'fossa subarcuata inferior.' Portman ('19 b), apparently without knowledge of Sterzi's work, suggested for the whole depression containing the saccus the term 'fossette endolymphatique' and stated that in about 60 per cent of cases the medial end is covered with a shell of bone from 1 to 3 mm. wide which conceals the true opening of the aqueduct. Not being aware of them, neither of these authors took into consideration the structural differentiation of the walls of the ductus and saccus. Certainly, the use of this aperture, according to the usual conception of it, as the boundary between parts given different basic names is, for the guinea-pig at least, a reversal of the general principles of descriptive terminology, although justified so long as nothing more essential was known. Therefore, it seems both proper and even necessary in the interests of clearness of diction in the future to attempt to name the parts of the membranous labyrinth concerned from this new viewpoint and in conformity with the general principles of terminology elsewhere. I realize fully the confusion which is apt to result from the introduction of new terms into a region already widely known by universally accepted terms, and therefore have decided that the use of modifications of the old terms with careful definitions is more to be desired than any selection of entirely new terms.

The primary division should undoubtedly be on the basis of the size of the lumen, as in other parts of the membranous labyrinth, and as was the intention of those who originally named these parts of it. The terms *sacculus*, *ductus endolymphaticus proprius*, and *saccus endolymphaticus proprius* are suggested for the primary divisions: the use of the modifying adjective 'proprius' being intended to distinguish this use of these terms as here defined from the previous one. The *ductus endolymphaticus proprius* may then be defined as consisting only of that part of the membranous labyrinth which has a relatively narrow elongated lumen; it connects the *sacculus* to the *saccus endolymphaticus proprius*; in the guinea-

pig all of its wall has a similar structure, and it extends from the apertura interna aquaeductus vestibuli to, on the average, somewhat beyond the middle of this bony canal. The saccus endolymphaticus proprius is then defined as all of that part of the membranous labyrinth distal to the end of the narrow lumen of the ductus endolymphaticus proprius; on the basis of the structure of its wall it may be subdivided into three portions, which may be termed for convenience the proximal, intermediate, and distal parts (*pars proximalis*, *pars intermedia*, and *pars distalis*). There is some reason to believe that such terms will be found applicable to many mammalian forms. The type structure of the wall for each of these parts in the guinea-pig is that shown in figures 5, 6, and 7, respectively, and described in the accompanying text matter (pp. 24 to 34). Attention is called to the fact that these terms as suggested and defined are not dependent upon the relations of the membranous labyrinth to the aquaeductus vestibuli, which appears to be of quite secondary importance, and variations in the relations to it do not affect in any way the applicability of these terms to other forms. In development the osseous formation is secondary to the membranous, and in the adult also it would appear that it should and, in fact, does continue to be of secondary importance. In the case of the guinea-pig, and this may prove to be quite different in some of the other species, the saccus endolymphaticus proprius is in such relation to the bony canal and to the dura mater that it is convenient to subdivide it also on this basis into two parts, which may be termed 'subosteal' and 'intradural.' Possibly 'intra-aquaeductular' is more proper than 'subosteal,' but it would at the same time imply that the term aquaeductus vestibuli should continue to apply to the whole length of this canal, and on this point it seems to me that the suggestions of both Sterzi and Portman are worthy of serious consideration, although in this discussion I shall continue to retain the BNA usage for the bony features. The term 'subosteal' simply indicates the position under cover of a bony wall, and the term 'intradural' is self-explanatory.

In the guinea-pig the pars proximalis of the structural basis is entirely subosteal on the basis of relations; the pars distalis is entirely intradural; while the pars intermedia is partially subosteal and partially intradural in position. In general, the pars proximalis forms distinctly the smallest part of the wall in the guinea-pig, while the pars intermedia and pars distalis seem of nearly equal extent so far as rough estimates go.

The guinea-pig has been so extensively used for the experimental work on both cochlear and vestibular parts of the labyrinth that it is surprising that the variations from the usual mammalian description given above have not been noted before. And yet the only account that I have found of this part in the guinea-pig in other than the embryonic stages is the three-page description by Portman ('19). That his description is very incomplete is evident from a comparison with the observations recorded above. It does not seem necessary to discuss all these points of difference in detail at this time, since most of them have been indicated in the course of the description. However, the question of the subepithelial connective-tissue spaces, which he describes as loosely joined so as to be continuous with the perilymphatic space, has not been referred to previously; my material shows many spaces in this loose areolar tissue, as in most such tissue, but I would hesitate very much to state that these are joined so as to form any continuous pathway communicating with the perilymphatic spaces of the vestibule. Sections alone are not sufficient to establish such a point, in my opinion. Siebenmann ('19) states very definitely for human material that these tissue spaces do not communicate with the perilymphatic space. In connection with the experimental work on the circulation of endolymph this question will have to be further considered.

For the bats which he studied, Iwata ('24) describes for the region of transition from ductus to saccus endolymphaticus a squamous epithelium about $2.2\ \mu$ thick with scattered $10\ \mu$ high cells projecting above the general level and resting

on the common basement membrane by means of $1\ \mu$ thick stalks between the squamous cells. These large cells become more numerous in the saccus proper and finally replace the lower cells and thus cease to project above the general level; Iwata says this epithelium has a maximum thickness of $11\ \mu$. Except for one part of the saccus, this forms the wall, and in certain parts he states that there are capillaries embedded so deeply into it that it looks much like stria vascularis. Of the one part Iwata says: "Hinten oben, schon in Bereich des eigentlichen Sinus, zeigt der Saccusrand ein ganz anderes Verhalten; das Epithel wird allmählich niedriger, erst kubisch, dann platt, letzteres besonders auf der Seite des Sinus. (Sinus transversus is referred to). Dann schwindet das Epithel vollständig, und das Lumen des Saccus endolymphaticus geht in die Spalten des hier lockeren Bindegewebes über." He then proceeds to speculate over the significance of this. One is reminded of Hasse's and of Rüdinger's ideas by Iwata's statement of a direct opening into tissue spaces, and I doubt if it will be confirmed by further observations; but his description of the presence of the columnar epithelium in the saccus endolymphaticus of the bat and of a rich vascular network closely related to this epithelium is in a general way in agreement with my findings in the guinea-pig and adds another mammalian form to the list of those in which the structure of the saccus differs from the usual account. No illustrations are given of this part of Iwata's work. It should be mentioned that the blood vessels in the guinea-pig are not in such close relation to the epithelium as described by him for the bat. The terms *pars proximalis*, *pars intermedia*, and *pars distalis* might well be applied to the description of the saccus of this animal. In general, the observations of Siebenmann ('19) on the human are also in agreement, so far as they go, with the essential structural differentiation of the walls of the ductus and saccus in the guinea-pig, although his report does not include all which one might expect his material to show. Of especial interest is the establishment in the adult human of an epithelium in the saccus which is dis-

tinety higher than the 'indifferente Labyrinthepithel' and like that which Boettcher ('69) described for the cat and the newborn human. His failure to find a basement membrane under this epithelium, in which his observations differ decidedly from those of Sterzi ('09), and especially the fact that to illustrate this point he selected for his sketches the region of the vascularized processes, indicate the possibility that the agreement in structure between the human and the guinea-pig extends even further.

It is of possible interest, from a comparative anatomical viewpoint, to note that in other classes of vertebrates also a columnar epithelium has been observed in the saccus endolymphaticus. In 1886, v. Lenhossék, in his work on the spinal ganglia of the frog, studied also the 'Kalksäckchen,' which were not then known to belong to the extensive saccus endolymphaticus of this form, and described for parts of these an epithelium 14 to 15 μ thick with numerous capillaries in the surrounding tissue, and he also mentioned being unable to locate any definite basement membrane under this epithelium. Rüdinger ('77) reported the enormous saccus endolymphaticus of the gecko (*Phyllodaetylus europaeus*) to be lined with a ciliated columnar epithelium. Wiedersheim ('76) had recorded this as squamous. And in *Acanthias vulgaris* and *Raja clavata* Retzius ('81) stated that the epithelium is columnar in the enlargement which in these forms with an external orifice is considered as homologous to the saccus in higher vertebrates. In view of the structures just described in the guinea-pig and of the functional significance discovered in the experimental part of our work, it would seem worth while to investigate the conditions in other forms also that the rather marked similarity of structural features gives a possible hint of.

The following quotation from Kolmer's ('23) account of the microscopic anatomy of the ear in the recent "Handbuch der Neurologie des Ohres," edited by Alexander, Marburg, and Brunner, represents very well the previous status of the knowledge concerning cellular elements in the membranous

labyrinth. He says (p. 121): "Irgendwelche Formen von Wanderzellen und weissen Blutelementen überhaupt scheinen im häutigen Labyrinth normalerweise niemals vorzukommen und dürfte deren Vorhandensein immer auf pathologische Vorgänge schliessen lassen." This one sentence is all that Kolmer has to say on the subject, and since for other topics he has included references to lower forms as well as higher, it would seem that if he were aware of any forms with such cells he would have mentioned it. So far as I am aware, my observations of cellular elements as normal contents of the saccus endolymphaticus were the first to be made on any mammalian form or, for that matter, on any animal. These cells were reported verbally in the presentation of my paper at the Buffalo meeting of the American Association of Anatomists, April 16 to 19, 1924, although they were not mentioned in the published abstract; and they were again reported upon at the Cleveland meeting, April 9 to 11, 1925, and at this time mentioned in the abstract. (Guild, '24, '25). Dr. Harold Cummins, after seeing my preparations (especially the ones from which figures 9 and 11 were sketched), examined his white rat preparations upon returning to Tulane University, and has written me, under date of September 6, 1924, that there are cellular elements normally present also in this form. He says, "Exactly the same variety of cell as you showed me in the guinea-pig material occurs here also." Concerning the more definitely blood-cell types he does not say anything. He further reports in his letter a 'coagulum, basophilic in reaction,' in which are many 'amorphous hyaline masses' which are further discussed in a second letter, but such masses I do not find in the guinea-pig; it may, however, be due to the effect of a different technique in preparation.

Siebenmann ('19) says, concerning the human material he used, that while the lumen is usually empty and collapsed, some cases had a content which in part took the nuclear stain; "derselbe ist meistens zurückzuführen auf amorphen feinen Kalkstaub, welcher in der Flüssigkeit suspendiert ist"; in

some there was also a mucous content and in one, a seventy-eight-year-old man, in this mucus-like content were relatively large masses of brownish-gray color with radiating striations. With the exception of these large masses, it would seem probable that the non-cellular content in the guinea-pig is much like that which Siebenmann saw in the human, although I have been unable to be certain that the dark-staining precipitate really contains any otolithic débris. One is reminded in this connection, as Siebenmann also was, of the conditions which have been known for a long time in many of the lower forms in which numerous otolithic crystals and débris are present in the saccus either during all or a part of the lifetime.

The normal presence of any type of content in the lumen of the saccus endolymphaticus and the absence of such content, whatever its nature, in the other parts of the membranous labyrinth are strongly indicative of a normal movement of fluid toward the saccus endolymphaticus; and although the possibility of local origin is not ruled out by this distribution alone, it is certainly very strong evidence against the normal movement of fluid away from the saccus toward the other parts of the labyrinth, as required by those theories which have been advanced in favor of the saccus endolymphaticus as the source of the endolymph. Since a separate account of our experimental work on the circulation of the endolymph is to follow this paper soon, the further consideration of the evidence afforded by the normal contents of the saccus will be postponed until it can be correlated with the evidence from the experiments.

In the tabulations on dimensions of parts other than those of epithelial tissue as such the weights of the animals have been included, and the tabulations have been arranged in the order of weight (tables 1, 2, 3). In all my records of material of this character I have watched for evidence of the correlation of the size of labyrinthine parts with the weight of the animals; as already stated, age data have not been available, so that weight is the best criterion of relative age

that I have. In view of the fact that in mammalia in general, so far as reasonably accurate data have been obtained, the growth of the labyrinthine parts is practically completed much sooner than most of the body, it was hardly to be expected that the size range in my series would show great differences, and the number cut in the exact plane that has made possible direct measurements is too small to generalize from. It has seemed worth while, however, to include the weights in the tabulations so that they will be available for comparison with other series which may each be too small in themselves for generalizations, but which when added to these may have greater significance. Without going into the details, since the number is too small to give any conclusive deductions, a study of the data when arranged and compared for the several elements shows that in general the average of the dimensions of the separate parts is greater in the heaviest group than in the lightest group, but the exceptions to this in individual animals are so numerous that it seems very possible that a longer series in each weight group might easily show any conclusions drawn from this series to be due to the effect of having wide individual variations from the mean in the few samples. Accordingly, I place the data on record as facts for these particular animals, but with the above word of warning as to attempting to draw deductions from them with respect to growth.

SUMMARY

1. The structure, the relations, and the normal contents of the ductus and saccus endolymphaticus of the guinea-pig have been studied in serial sections. In eleven of these series the orientation was such that a single section included the whole length of the lumen of the ductus endolymphaticus. On these direct measurements of certain dimensions have been made. Particular attention has been devoted to the details of the histologic structure of the walls of these parts and a comparison made with that of the sacculus, to which they are commonly stated to be similar. On the basis of mor-

phology alone, three very distinct types of wall have been located and described; each is different from that of the sacculus.

2. The inadequacy of the terms ductus and saccus endolymphaticus for the designation of the parts described and the lack of correlation of the conventional boundary between these with the more essential morphologic facts have been indicated. Modifications of the old terms are suggested. It is suggested that the term ductus endolymphaticus proprius be applied to only the elongated narrow portion of the membranous labyrinth extending from the sacculus through the proximal part (slightly more than half of the length in the guinea-pig) of the aquaeductus vestibuli; and that the term saccus endolymphaticus proprius be used for all of the part distal to the beginning of the funnel-like expansion, irrespective of its relation to the bony canal. The wall of the ductus endolymphaticus proprius in the guinea-pig is formed of simple squamous epithelium with scattered basal processes and with simple pointed crypts extending into a very scantily vascularized loose areolar tissue. The transition between wall of this structure and that typical of the sacculus coincides with the end of the narrow lumen and also with the region of the internal aperture of the aquaeductus vestibuli, and this is about 0.3 mm. distant from the saccular end of the utriculo-saccular duct. These facts are considered as additional evidence on the question of the proper terminology in this region.

On the basis of the three types of wall described for the saccus endolymphaticus proprius, it is suggested that for descriptive purposes it be subdivided into three parts, to be termed *pars proximalis*, *pars intermedia*, and *pars distalis*. There is reason to believe that such a terminology is applicable to many mammalian forms. For the guinea-pig the parts may be briefly located and characterized as follows: *a*) The *pars proximalis* lies entirely within the dilated portion of the aquaeductus vestibuli and most of its wall is similar to that of the ductus, but a region of transition with gradually increasing thickness of the epithelium is considered as belong-

ing also to this part. *b*) The *pars intermedia* lies partly within the aquaeductus vestibuli and partly between the layers of the dura mater near its external or cranial aperture, extending posteriorly to the general region of the anterior margin of the transverse venous dural sinus; its wall is characterized by a predominance of a columnar epithelium which for the most part does not have any distinct basement membrane; this part has both crypts into the surrounding richly vascularized areolar tissue and epithelial processes into the lumen, some of which are and some of which are not vascularized. It is not only structurally a specialized area, but is also specialized functionally, as has been shown in the experimental work reported at the Buffalo meeting of the American Association of Anatomists and soon to be published in a separate account. *c*) The *pars distalis* overlies the transverse sinus in the dura mater, and its lumen, as seen in sections, may be patent or may be merely potential; its wall is composed of a thick squamous epithelium resting on a relatively dense areolar tissue which contains only a few small blood vessels. Occasionally this part extends slightly beyond the transverse sinus, and in some of these the wall of this extreme end resembles more that of the intermediate part. On the basis of the relations of the saccus endolymphaticus proprius to the aquaeductus vestibuli, the division into parts termed 'subosteal' and 'intradural' is suggested as convenient for the guinea-pig.

3. Numerous free cellular elements are normally present in the lumen of the saccus endolymphaticus of the guinea-pig. Drawings of many of these cells are given, but, because of the limitations of interpretation based solely upon fixed and sectioned material, only tentative classifications have been made of most of these. Some belong undoubtedly to the group of white blood cells; many are definitely phagocytic in character; many are of quite large size and some show evidence of having possessed the power of ameboid movement. Two types of precipitate are present in the lumen of the saccus endolymphaticus in the sections, one of these stains

very heavily with iron-lac hematoxylin. The functional significance of the normal presence in this part of contents different in character than found elsewhere in the membranous labyrinth is briefly indicated.

4. The weights of the guinea-pigs used in the measurements are placed on record in the tabulations, but the evidence of definite growth of the parts concerned in the period covered by the series is inconclusive.

5. The literature on the structure and contents of the ductus and saccus endolymphaticus in all mammalian forms for stages later than the embryonic has been carefully reviewed because of the absence in other recent work of a review to which reference could be made for those interested.

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