

THE FUNCTIONAL DEVELOPMENT OF THE MESONEPHROS
IN PEROMYSCUS LEUCOPUS

by

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THE FUNCTIONAL DEVELOPMENT OF THE MESONEPHROS IN
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O. Hertwig in his text, *Elemente der Entwicklungslehre*, introduces the topic of the development of the urogenital system with the statement: "We turn herewith to one of the most interesting sections of Embryology, since here, as with no other organ system are effected many important metamorphoses during embryonic life." Felix, in Keibel and Mall's text book of Embryology continues the same thought. "We have to deal, not with the gradual development of a single organ, which is laid down and brought step by step to its definitive completion, but with a saltatory development; three organs (pronephros, mesonephros, and metanephros) are developed in succession, and each of these organs is apportioned and adapted to a definite period of one entire development."

This field for investigation has appealed to many workers. The literature is voluminous yet there are many questions still unanswered and many details which have not been worked out.

The purpose of this paper is to give a detailed

account of the development of the mesonephros and its relationship to the pronephros and metanephros in the deer-mouse, Peromyscus leucopus. This organ was found to be very degenerate with no histological indications of functional activity. No glomeruli are developed in the tubules, excepting for very rudimentary ones in the posterior end during the later stages of development of the organ. The circulation of the organ is apparently entirely venous.

The literature divides itself in general into three groups:

1. Those papers dealing with the phylogenetic development of the urogenital system.

2. Those which concern themselves with the ontogenetic development and morphological relationships of the parts of the system.

3. Papers investigating the functional development and the physiological activity of the various organs.

- Balfour (1875) studied the development of the mesonephros in the selacians and the amphibia and to a lesser extent in the cyclostomes, the ganoid and the bony fishes.

In the selacians, from the intermediate cell mass opposite the fifth somite a solid knob develops from which a column of cells grows back to opposite the position

of the future anus. The knob projects outward and toward the ectoderm and the column lies at first between the ectoderm and the mesoblast. The knob then becomes hollow and acquires a wide opening into the pleuroperitoneal cavity. The column of cells becomes a hollow tube and soon opens into the cloaca. In the meantime solid outgrowths of the intermediate cell mass appear at the uppermost corner of the body cavity. They soon become hollow and join the long segmental duct. One tubule develops for each somite. Each opens into the coelom. These tubules are mesonephric.

Now a longitudinal division of the segmental duct into the Wolffian and Müllerian ducts occurs in the females and in the anterior end, at least, in the males.

Later one or more other ducts form from the posterior portion of the Wolffian duct which receive the secretion of the posterior segmental tubules. Balfour considered these ducts and the tubules which empty into them as homologous with the ureter and metanephros of higher vertebrates. He considered that originally each collecting tubule of the ancestral kidney opened to the outside independent of the others in a manner probably similar to the nephridia of the annelids. The Wolffian duct represents the first of these collecting tubules

and a possible fusion and suppression of the rest.

After making comparisons of the development of the mesonephros in the other forms he concludes that the mesonephros of the cyclostome, of bony fishes and in the majority of points of the amphibia, are more primitive and exhibit a decidedly less differentiated condition than in the selacians.

He lists five differences between the amphibia and the selacians.

1. The amphibia have a functional pronephros. It is suppressed and non-functional in the selacians.

2. There is a less complete separation of the Wolffian and Mullerian ducts in the amphibia than in the selacians.

3. The selacians differentiate the mesonephros into two glands comparable to the mesonephros and metanephros of higher forms. The amphibia have only the mesonephros.

4. The Mullerian duct persists entire in the male amphibia while only the upper end is retained by the selacians.

5. The tubules in the selacians are segmental while in the amphibia they are numerous and branched in each segment. This is the only point in which the amphibia show greater differentiation than the selacians.

Hertwig (1910) also describes the development of the selacian pronephros. He finds that in the place where the segmented part of the middle germ layer continues over into the unsegmented part a number of segmental cell cords grow out of the parietal layer which bend back toward the posterior and unite to form a longitudinal cord. These cell cords then acquire cavities which communicate with the body cavity at one end and the cavity of the pronephric duct at the other. Shortly after its formation the anterior half of the organ completely degenerates while the posterior half develops further and enlarges, but remains in connection with the body cavity through a single funnel only. As to the status of this one funnel he does not commit himself but points out that van Wijhe considers this funnel to be the result of the fusion of several tubules into one, while Rückert considers that all of the funnels with the exception of one close themselves and degenerate.

In discussing the origin of the Wolffian duct Hertwig (1910) arrives at the conclusion (and states that this is also the opinion of Felix) that among all vertebrates with the exception of amphioxus, the anterior portion of the pronephric duct develops from the middle germ layer as a result of the fusion of the free end of each pronephric

tubule with the tubule next posterior to it. This anterior region of the duct ends with the formation of the last pronephric tubule.

The posterior portion of the duct is formed in one or the other of two ways. In bony fishes, amphibia, reptiles and birds the terminal portion of the anterior region is the free backward projecting protuberance of the last pronephric tubule. This protuberance then grows gradually in length through proliferation of its own cells until it reaches the cloacal wall with which it fuses. He emphasizes, then, that the middle and hinder region of the pronephric duct in these classes of vertebrates does not constrict itself off from either the outer germ layer or the middle germ layer nor does it derive any material from either of these layers for its enlargement.

On the other hand, in the mammals he considers that the protuberance at the posterior end of the anterior region, which has arisen from the middle germ layer, instead of ending free as a knob, places itself immediately in solid union with the outer germ layer. A ridge on the outer germ layer then grows posteriorly from the end of this protuberance. As the ridge lengthens posteriorly it gradually detaches itself from the outer germ layer, beginning at the anterior end so that it is only united

with the outer layer at the posterior or growing end of the ridge. In defense of this opinion he cites Hensen, Fleming, Graf Spee and Keibel as agreeing with him.

Kerns (1907) found the pronephric duct in the mole closely applied to the ectoderm but found nothing to indicate that it was derived from the ectoderm.

In Peromyscus leucopus the same condition is found. Although the duct in its formative stage is in close contact with the ectoderm throughout its length it is always possible to distinguish the line of separation between the two. The duct develops very rapidly and reaches the region of the cloaca while it is still in the form of a solid cord. It does not, however, fuse with the wall of the cloaca at this time. One noticeable fact in connection with the development of this duct is the relative absence of mitotic figures in the cells either at the tip or in other regions.

We return now to Balfour's conclusion that the selacians are phylogenetically more advanced than the cyclostomes, teleosts or amphibia in so far as the development of the kidney is concerned. In the following extract quoted from Kiebel and Mall (P. 867), discussing the phylogenetic development of the metanephros, Felix describes embryonic metanephroi in the gymniophiones,

birds and in man which are quite similar to the organs described by Balfour in the selacians.

"We must define again in all clearness what we mean by the metanephros. It is an excretory organ whose tubules open into special collecting tubules, developed from the primary excretory duct. Whether these special collecting tubules are represented by a simple tube or develop to such an expanded ureteric tree as we see in man, the opening of the metanephric tubules into a special efferent duct developed from the primary excretory duct is the characteristic feature.

"Starting with this point if we consider, first of all, the development of the mesonephros of the Gymnophiones, we will find that we have to distinguish in it between ventral mesonephric tubules, all of which are in a longitudinal row at the summit of the mesonephric fold and dorsal mesonephric tubules which lie at the foot of the fold and, in order to open into the primary excretory duct, must traverse a much longer path than the ventral ones. These dorsal tubules are developed for the most part later than the others and may occur in varying number and different series. The later formed series are always the more dorsal ones, and, finally, their tubules can no longer reach the primary excretory duct

by their own efforts. Accordingly the primary excretory duct by their own efforts. Accordingly the primary duct sends out special collecting tubules for the reception of these canals; according to our definition we would have to regard these dorsal mesonephric tubules with their special collecting tubules as metanephric structures. In the mesonephros of young Gymnophiones, then, there is scattered throughout its entire length a series of metanephroi; only the caudal ones persist, however, in the adult. In birds a similar observation is to be recorded. It must be pointed out, in the first place, that the length of the mesonephros in birds is markedly less than that of the mesonephros of the Gymnophiones, and, consequently, the mesonephric tubules are compressed into a smaller space. What the mesonephros loses in length, it gains in thickness, and since a great amount of development is possible only in the caudal portion of the body cavity, the mesonephros increases in man- i.e., develops dorsal tubules- principally in this region. These dorsal tubules are too far away to open directly into the primary excretory duct, and, therefore, special collecting tubules are formed for them by the duct. According to our definition these dorsal mesonephric tubules, together with their collecting tubules, are also metanephroid.

Their number, however, for the reason stated, is small; we have only a few metanephroi and a few special collecting tubules.

"Finally, we have shown that in the development of the human mesonephros a heaping up of tubules occurs at the caudal end; furthermore, that these mesonephric tubules no longer lie in one series, and, finally, that special collecting tubules are formed for them from the primary excretory duct, but these are few in number in comparison with the condition in the chick.

"In brief, then: At first numerous metanephroi are interposed between the mesonephric tubules; the more the space in the body cavity becomes limited as development proceeds, the more also the number of these metanephroi becomes diminished, until, finally, in adult mammals only one is developed.....

"I have finally to consider why the last ureter becomes the metanephric ureter. I would associate this with the preference which the last ureter acquires in reptiles and birds. In these the mass of the mesonephric tubules extends caudally beyond the primary excretory duct and forms the so-called caudal kidney. For the tubules of this the primary excretory duct forms a special strong ureter, which must be the last of all these ureters. By the development of the

caudal kidney the last ureter acquires a special importance and therefore a preference which finally allows it to dominate over the others as the metanephric ureter."

If these interpretations are correct we are led to consider the metanephros as it is found in the adult amniotes as the more highly developed expression of an organ which has existed phylogenetically throughout all of the vertebrate history. We will return to this topic later under the discussion of Schreiner's work.

The question of the relationship which exists between the pronephros, mesonephros and metanephros in the amniotes has been the object of numerous papers and has resulted in at least three interpretations. Sedgwick and Balfour (fide Kerens) expressed the opinion that the pronephros, mesonephros and metanephros were only different parts of one and the same organ appearing in different places during the successive stages of development. Price (1897) has applied to this organ the term 'holonephros'. Felix (1912), although he does not agree with this interpretation of the relationships, makes the following statement (P. 755). "Each primitive segment stalk may develop several tubules in succession, since it itself passes along a definite path of development, during which its form changes, and opportunity is afforded for differ-

ences between the earlier and the later formed uriniferous tubules. Pronephric and mesonephric tubules of the same animal need not therefore show a similar development, if, between their formations there occurs a distinct interval, during which their common source has acquired a new form. Similarly the pronephric tubules of different animals show profound differences in their development, differences which find an explanation in the time relations of the formation of the tubules. The change of form which accompanies the progressive development of the common source of the organ suffices to explain the difference in development of a pronephric, mesonephric and metanephric tubule." Brauer (1902) has made a very careful study of the development of the excretory system in the gymniophione, Hypogeophys. Among the gymniophiones probably a more complete and diagrammatic development of these organs in their primitive form is to be found than in any other vertebrate, and this in spite of the high degree of specialization of the animals themselves. Brauer's general conclusion concerning the relationship of the pronephros and mesonephros is this: Pronephros and mesonephros are parts of equal significance, developing at different times and places, of one and the same apparatus extending throughout the trunk. The only differences between the pronephros and mesonephros, as he

points out, are of secondary importance; for example: the relations of the tubules with the excretory canals; the mesonephric tubules only terminating secondarily in a canal already formed. But he finds that most of the pronephric tubules do the same thing since only the first three contribute to the formation of the pronephric duct.

The second opinion as to the relationships of the pronephros, mesonephros and metanephros was advanced by Gegenbaur (fide Kerens) and others. They considered the three organs as essentially different; that they have succeeded each other phylogenetically in the same order in which they develop ontogenetically; that the mesonephros replaces in all its parts the vanished pronephros and that in turn it is completely replaced by the metanephros. The general impression concerning this viewpoint seems to be that it is the result of a morphological study of the individual organs rather than of a systematic study of the embryonic history of the organs.

The third opinion was advanced by R^uckert (1892) and has had a strong following among many of the leading authors. R^uckert considers the pronephros as a complete organ while the mesonephros and metanephros appeared later as a second and third generation of tubules both derived from the same portion of the nephrotome but more dorsal than that from

which the pronephros was derived. He considers that the primitive excretory organ of the actual ancestors of the vertebrates extended the entire length of the trunk consisting of a pronephros formed of segmental tubules, diverticula of the coelome and each opening independently to the outside. Later the distal extremities of these canals fused into one collecting canal which alone extended to the surface of the body. Subsequently all of the middle and posterior part of the organ atrophied with the exception of the excretory duct. After this atrophy, in a second growth, some new tubules develop arising from the mesoblast situated dorsally with relation to that which had furnished the tubules of the pronephros and these tubules of the second generation coming themselves to open into the excretory canal of the pronephros constitute the mesonephros. Later still but from the same region of the mesoblast as the mesonephros, a third generation of tubules arose. These, because of their greater distance from the excretory canal, could not open into it and as a result collecting canals were evaginated from the excretory canal to meet these tubules. Later still all of these third generation tubules disappeared in the adult animals excepting for those most posterior which persist as the metanephros. During embryonic development of many forms however they are still present.

Felix agrees with the opinion of Ruckert. The following paragraph is quoted, (Keibel and Mall P.798).

"In those vertebrates in which the pronephros extends the entire length of the body cavity, it is at once evident that the pronephric tubules form a first and the mesonephric a second generation of excretory tubules. The mesonephric tubules are, accordingly, something new, distinguishable from the pronephric tubules; it will occur to no one to regard the fruit of one year as of the same generation as that of a preceding year. In the vertebrates in which the pronephros is shortened, the matter is not so clear; indeed, it may be so obscure that one is led so far astray as to regard the pronephric and mesonephric tubules as equivalent parts of a single system. All investigations that have led to this estimate of the mesonephric tubules have been made on vertebrates with a shortened pronephros. Whoever examines the development of the pronephros and mesonephros throughout the whole series of vertebrates cannot for a moment doubt that the pronephric and mesonephric tubules are members of two different systems."

Felix points out the characteristic differences between the pronephric and mesonephric tubules and then goes to some length in explaining how these differences

all have their exceptions. The formation of the tubule before the excretory duct is formed is the only real criterion for determining a pronephric tubule just as the formation of the tubule and its later joining the already formed excretory duct is his criterion for determining a mesonephric tubule in those regions where either sort might be found. He states that both sorts of tubules may be found in the same segment. Quoting him on this point: "If a pronephric and a mesonephric tubule are formed in the same segment and if, furthermore, the anlage of the mesonephric tubule appears while the pronephric tubule is still present, then it is possible to distinguish the two by their position, the pronephric tubule lying lateral nearer to the lateral plate, the mesonephric one medial, nearer to the primitive segment. But whoever has studied the development of the two provisional organs in the vertebrate series must admit that these two conditions are very rarely fulfilled."

According to Brauer (1902) the rudimentary mesonephric formation which Felix describes are the remains of the union of the nephrotome with the scleromyotome.

Since the chief point of difference between the various authors has been the relationship between the pronephros and the mesonephros Kerens (1907) has made

an extensive study of these relationships as they appear to exist throughout a series of forms representing each of the groups of amniotes accepting Brauer's work on the gymniphones as conclusive for the anamniotes. She studied very complete series of embryos from the first phases of the appearance of the excretory organs up to the full development of the mesonephros. Of the reptiles she studied, Lacerta muralis, L. agilis and Tropidonotus matrix. Of the birds the chick embryos and of the mammals the embryos of the mole and the rabbit. Of this last group she did not have a complete series of the mole. However, the series of the rabbit overlapped the other to such an extent that detailed comparisons were possible and the structures were found to be so nearly identical that the rabbit embryos were used to fill the gap. She has described segment by segment the development in the embryos of these various forms. The details or even the points which she stressed cannot be revealed here. Suffice to say that her final conclusion is that the pronephros is only the anterior extremity of the mesonephros.

Schreiner (1902) has made almost as exhaustive a study of the relationships existing between the mesonephros and metanephros. His studies were based on embryos of Lacerta agilis, the chick, the rabbit, the pig and man. He appears

to have demonstrated very clearly the morphological identity between the mesonephros and the metanephros.

The majority of the authors have concerned themselves with the phylogenetic relationships of the various parts of the urogenital system, with the origin and relationships of the various ducts or parts of the system, or with the general anatomy. Only a few papers discussing the mesonephros have described in more than generalized terms the essential tissues, i.e., the tubules, of this organ. Among those authors making a detailed study of the tubules are Kollmann (1898), Felix (1912), Kölliker (1897), Mihalkovics (1885), Mac Callum (1902) and Lewis (1920). Kollmann modeled a tubule from the mesonephros of a 10.2 mm human embryo. This illustration has been reproduced many times. Felix (Keibel and Mall) has given detailed descriptions of the tubules as they occur in human embryos. Concerning his work, However, we quote from Lewis (1920). "But Felix records that he has 'studied two hundred models of mesonephric tubules from the most different stages of development', and in none of them has he found 'any coiling of the tubule'. Consequently he infers that Kollmann's model must represent 'a very exceptional rarity' - a conservative and accurate conclusion. The only change observed by

Felix after the tubule has become S-shaped occurs near the junction of the middle and distal segments of the S (distal meaning toward the Wolffian duct), and consists in the formation of 'a loop directed either cranially or caudally'. Unfortunately Felix has not figured in detail any of the late stages included among the two hundred tubules modeled, presumably from human embryos, so that Kollmann's drawing appears to occupy the field alone."

Kölliker, in 1897, according to Lewis probably described for the first time the S-shaped stage. His study was based on rabbit embryos. He writes: "From the Wolffian duct there arises first a very slender tubule which passes medially along the dorsal side of the Wolffian body, clear across the organ; then, making a loop, it bends upon itself and retraces its course to the lateral side; finally, after a third coil, it ends in the Malpighian corpuscle, medially placed on the ventral side." He disposes of the later stages as follows: These three chief coils become complicated by the formation, at the places where the bends occur, of accessory coils in different planes, so that finally the course of a single tubule becomes so complicated that it cannot be unravelled in sections."

Mihalkovics (1885) made studies on the development of the mesonephric tubules in lizard, the duck, the chick and the sheep. He describes the tubule as beginning as a detached vesicle which elongates into a tubule. This becomes cupped or crescentic with the glomerulus developing in its concavity. As it joins the Wolffian duct it assumes a "sickle-form" and from this the S-form is developed. Later, in the sheep, he found that the middle or transverse limb of the S elongates and becomes itself S-shaped, though the long axis of the second S is horizontal, being at right angles with that of the first resulting in a double or reversed spiral form for the tubule. He did not follow the development of the tubule further.

Schreiner (1902) in his work embodying a study of the lizard, duck, chick and rabbit, refers to the reversed spiral as a 'much coiled' S.

MacCallum (1902) has made a detailed study of the mesonephric tubules in the pig embryo and has made wax reconstructions of them. The gland and its tubules attains a greater development in the pig than in any other mammal for which the details have been worked out. Because of this greater development the description of the tubule as given by MacCallum will be quoted to

facilitate comparisons with those forms with a lesser developed mesonephros. "The course of the tubule can be made out plainly from its beginning in the Malpighian body to its termination in the Wolffian duct. The Bowman's capsule (to use a term usually employed in describing a similar structure in the permanent kidney) narrows down to a fine tube which runs forward toward the ventral border. Here it turns and follows the lateral surface of the gland to a short distance from the dorsal border, where it turns abruptly on itself, forming a large loop, and returns to the region of the anterior border, Here it becomes somewhat convoluted and then passes over to the region of the dorsal border, where it is again thrown into convolutions. From the dorsal border it proceeds around on the lateral surface of the gland to empty into the Wolffian duct. Certain differences in the calibre of the tubule are to be noted. The collecting tubule arising in the capsule of Bowman is small, and is lined by cubical epithelium. In the region of the lateral surface it passes into a tube many times larger, lined by large columnar epithelial cells containing granular protoplasm. These cells seem to be secretory in character. This large tube forms a complete loop, and passes over in the region of the

anterior border into a much smaller, somewhat convoluted segment of the tubule. This in turn runs across the surface of the Malpighian bodies, where it becomes again greater in diameter, to join with another convoluted segment in the region of the dorsal border. This whole middle part of the tubule has a much greater diameter than either the collecting tubule at the glomerulus end or that which empties into the Wolffian duct." "There is, however, a very distinct division into a secretory and a conducting part."

Lewis (1920) used the figures of the modeled tubules published by MacCallum, together with a series of human embryos from which he reconstructed the tubules, as the basis for a paper in which he attempts to assign a more or less definite basic pattern to which the tubules of the mesonephros may all be reduced; some, as in the pig, by elaboration; others, as in man, by simplification. He takes as his simple pattern the double spiral tubule which has developed from the S-shape. He divides the tubule into three parts the divisions having reference to the form of the portion of the tube and not to the possible physiological divisions. The portion beginning at the capsule and forming the first loop he designates as C. The next also because of its shape he names U and finally the last

loop which connects with the Wolffian duct and which also has a 'simple C-shaped curve' he calls Z. The tubule of the pig, which at first sight seems quite complex, is reduced to the simple pattern by applying the divisions suggested by Lewis and noting that secondary coils have appeared in the sides of the U portion. Concerning the model of a fully developed tubule from a human embryo he says: "Although the tubule at this stage is of a simpler pattern than the double spiral, and consists of two rather than three segments, the remains of the middle or U-portion can be identified. A narrowing of the tubule marks the place where the U begins and it proceeds around the bend into Z, where it ends without definite boundary."

Huber (1917) has prepared a model of the tubule of the frog which can, according to Lewis, be reduced into its general aspects to the same pattern as described above.

Although the number of forms which have been studied are too limited to make a definite statement it seems probable that the tubules of the mesonephros can be reduced to a general pattern as has been done for the tubules of the metanephros.

Comparatively little attention has been given to the

functional development of the mesonephros among the Amniotes. Weber (1897) was probably one of the first to attempt to find out whether the mesonephros actually functions as a kidney during embryonic life. He examined series of embryos of the pig, the mole, the guinea pig, the mouse and man. In the pig, as is well known, he found a large mesonephros, which remained in a condition capable of functioning as a kidney until after the metanephros was sufficiently developed to be able to take over its function. In the mole and the guinea pig the mesonephros is small and temporary, that is, its degeneration beyond the stage of possible functional activity occurs before the metanephros is sufficiently developed to take over the work of excretion. In man the same condition exists, while in the mouse the organ is still more degenerate, never developing glomeruli and degenerating very early. Considering the allantois as the receptacle for the mesonephric urine he studies its development along with the development of the mesonephros. He finds in some rodents that it never exists as a sac outside of the body cavity and in man is only developed as a slender tube extending into the umbilicus. If the mesonephros were functional then in these forms there was no reservoir for the storage of the excretory products. He also gave

considerable attention to the time when the urogenital sinus opened but found that it did not open until well after the peak of development of the mesonephros had been reached and accordingly there was no possibility of the urine being discharged from the cloaca into the amniotic cavity. He concluded that in those cases where the mesonephros degenerated before the metanephros could have begun its activity, that the mesonephros did not function, "for if it had been active and had then degenerated before the metanephros had begun its activity, there must have been a certain period of development during which there was no excretion."

This argument was apparently satisfactory to all for it was not questioned for over twenty years. That Felix accepted it is shown by the following statement: "In an embryo 22 mm greatest length none of the mesonephric tubules were capable of functioning; in all the tubulus secretorius had separated from the tubulus collectivus. If one inquires how far the development of the metanephros has progressed at this time one finds that embryos of 22 mm. have just reached the anlage of the 2nd generation of uriniferous tubules. The first generation however, has as yet no fully formed Malpighian corpuscles. If, then the mesonephros has functioned as an excretory organ, there

must necessarily have been an interruption of this function on its degeneration. Consequently I regard this question as to the functioning of the mesonephros as settled; it does not function as an excretory organ. This does not, of course, imply that it may not have been active in another manner unknown to us."

Bremer (1902) considers that both Weber and Felix from their assumption that the mesonephros is not functional in some mammals also assume that it is not functional in any mammals, and also, that since they assume that the mesonephros is not functional, then, there is no excretion during embryonic life. He says: "The possibility that some other fetal organ might assume the excretory function during the interim when neither the mesonephros nor the kidney is apparently capable of activity, or might even in some animals like the rat and the mouse replace the Wolffian body partially or entirely, is not considered by either Weber or Felix; yet that this is the case, and that this organ is the placenta seems to me to be strongly suggested by the facts brought out in this paper."

He finds in the placentae of certain animals cells which so closely resemble the essential cells of the Bowman's capsule that he assumes that they are identical both in structure and function and that they form a

placental excretory organ "capable of serving the fetus when neither the Wolffian body nor the kidney is active."

Further, he points out that this placental excretory organ is not as well developed in some animals as in others and that the extent of development is more or less correlated with the stage of development of the embryonic kidney and the allantois.

The cells which are the essential part of the glomerulus, whether of the mesonephros or of the metanephros, in their mature form are flattened, having an "extremely thin, flange-like process, extending from one side of the cell, while the remainder of the cell, including the nucleus, retains its cuboidal shape." The flanges of these cells fuse with each other so that the cell limits cannot be recognized. The blood vessels of the glomerulus are covered by these thin plates and it is through them that the fluids from the blood enters the tubules.

Bremer counted and measured the glomeruli in the embryos of the different mammals studied in order to have a definite idea as to the relative length of time during which the Wolffian body, or at least the part of it represented by the glomeruli, may be considered functional.

At one end of the scale he placed the rat and mouse

which never develop glomeruli, next to these come the guinea pig in which the glomeruli are never large or numerous and, also, from the observations of other authors, he places the mole in the same class. He doubts if the mesonephros is ever functional in these forms. In the rabbit the mesonephros develops early into a large organ with large glomerule, but its atrophy is also early. In man there is a small Wolffian body, early developed to its full capacity but only retaining its function as far as the glomeruli are concerned until the second or third month of intra-uterine life.

In sharp contrast to these are those of the pig, sheep and cat. In the pig it seems certain that the activity of the mesonephros overlaps that of the metanephros. In the sheep, in spite of certain variations in structure in the corpuscles, there is a large number of functional glomeruli which persist for a considerable time until the kidney is capable of functioning. In the cat the glomeruli are small but persist until after the kidney is functional.

The allantois, he finds to be quite large in the pig and sheep, smaller in the cat, and very small, limited to the area of the placenta, in the rabbit. In man and the guinea pig it only develops as an allantoic

stalk which may or may not have a cavity. In the rat and mouse no allantois is developed. Quoting him: "It will be seen at once that in the animals selected for study there is a close relationship between the size and duration of the Wolffian body and the size of the allantois. The cat has a smaller allantois than the pig or sheep, because its Wolffian body is less effective, but a larger one than the rabbit, since the urine is accumulated throughout life in the cat but only for a short period in the rabbit."

From a study of the placenta of these different forms he finds "that the placentae of all of that class of embryos in which we have found an early involution of the mesonephros exhibit membranous plates in proper relation to the fetal vessels, and that the time of the disappearance of the one is approximately the same as that of the appearance of the other. Moreover, the placentae of those animals studied which are able to utilize the Wolffian body until the kidney is ready for action show no such modification of the ectoderm."

Finally he concludes that "it appears that embryonic and fetal urinary excretion takes place wholly through the placenta in the rat, at first through the Wolffian body and later through the placenta in the rabbit,

guinea pig and man, but never through the placenta in the pig, sheep or cat."

To substantiate his contention that the embryonic excretion can be carried on in the absence of a functional kidney he cites cases reported by English of the obliteration or stenosis of the urinary passages in fetuses and in the new born. Usually death occurs in the sixth to the eighth month, but in other cases, the child was born healthy and showed uremic symptoms only after two or three days.

Further evidence of the taking over of the function of the kidneys by other organs of the body is found in the work of Smith(1929) on fishes. Because of the very small total nitrogen excretion by way of the urine in various fishes studied by him, Smith was led to suspect that the nitrogen, possibly as ammonis or urea, was being lost from the body by some route other than the kidneys and since the gills appeared to offer the most probable avenue of escape he carried out a number of experiments to determine if this was the case.

For the tests two species of carp were used, Cyprinus carpio and Carassius auratus. Holes were cut in two strong rubber dams (two being used to lessen leakage) which were then fastened between the cut ends of a box. The

fish were slipped head first through the holes in the rubber dams so that they closed around the body just back of the pectoral fins. Thus the excretion from the gills was kept separate from any which might be given off from the skin or kidneys. As a further check the urine from the kidneys was collected by a glass retention catheter sewed in the urinary papilla and emptying into a small rubber bag which was fastened to the anal fin. Fish were kept in the box for as much as twenty four hours.

Within ten minutes after putting water on the fish he demonstrated ammonia in constantly increasing quantities in the front chamber of the box while none appeared in the rear chamber for several hours. Further the excretory roll of the gills is substantiated by the finding of urea in the water removed from the front chamber.

From many other experimental data it has been shown that other organs of the body than the kidneys are capable of secreting and excreting the products of nitrogenous metabolism.

In so far as is known at present, certain Mediterranean fishes are the only other vertebrates, besides the rodents discussed in this paper, in which a kidney is

developed which does not have glomeruli. Edwards (1928a, 1928b, 1929) has recently made some very interesting and quite extensive studies on the kidneys of these fishes. The points of his results which have a possible relationship to this paper are quoted from his summary.

"The aglomerular tubule differs from the glomerular tubule of fish, amphibia, and reptilia in lacking Bowman's capsule and an intermediate segment and, therefore, in the cytological modifications characteristic of these regions.

"Despite the fact that the morphological and cytological differentiation is less in the tubules of aglomerular mesonephroi, the urine eliminated by such mesonephroi is comparable to that of the higher vertebrates, including man. From this the conclusion is drawn that the tubule is the more important structure, phylogenetically and functionally."

MATERIALS AND METHODS

The embryos for this study were obtained from females caught in the field and killed immediately and from mice reared in the laboratory. Both sources proved to be very time consuming and wasteful of mice.

As mentioned in another paper (McNair, 1930), although *Peromyscus leucopus* breeds prolifically in captivity it was found to be impossible to time the breeding and to get embryos of a definitely known age.

Brambell (1927) studied the morphology and development of the gonads of the mouse, (presumably the white mouse commonly used in the laboratory). He was able to get fairly accurate data as to the age of the embryos by "calculating from the date of finding the plug." This procedure was impossible with the deer mice. Handling them each day to examine them for the plug disturbed them so much that they refused to breed at all. The descriptions and illustrations in Brambell's paper do not furnish any information which could be used to determine even approximately the age of the embryos used for this study.

As apparently an only means of identifying the

embryos with any degree of accuracy a camera lucida drawing was made of each one and also from the same level as the embryo a millimeter scale was copied beside the drawing of the embryo. As can be seen in the illustrations a statement of any of the customary embryonic measurement is of little value because of the varying degrees of coiling of the different embryos.

When a female containing embryos was killed the uterus was removed and transferred to warm normal saline solution where in the majority of cases the embryos were removed from the uterus before placing them in the fixing fluid.

For fixing agents, Gilson's fluid, Bouin's fluid and Formal-alcohol-acetic solutions were used. The best results were obtained from those embryos fixed in Gilson's fluid.

After dehydration the larger embryos were cleared in cedar oil, washed in xylol and infiltrated for varying lengths of time in a mixture of xylol and paraffin according to the procedure described by Malumphry (1928). The very small embryos were simply cleared in xylol. Some of these were removed from the uterus but the best results were obtained by removing only enough of the ureine wall after dehydration to make it possible to see

the position of the embryo for orientation purposes.

All embryos were embedded in paraffin and sectioned at 10 microns. Embryos of each stage were cut in both cross and longitudinal sections. Owing to the small size of the mesonephros and to the almost horizontal direction of the tubules with reference to the long axis of the organ embryos cut in longitudinal section were of comparatively little value.

All of the sections were stained with Delafield's hematoxylin and eosin.

All drawings were made with the camera lucida. The drawings for the wax reconstructions of the tubules, Figs. 22, 23 and 24, were made at a magnification of 1000 diameters. The other drawings were made at the magnification which best brought out the desired details.

For the reconstructions, each section was carefully outlined on paper, transferred to sheets of wax which had been rolled to the proper thickness and cut out. The wax plates were then stacked, pinned together and smoothed off before the drawings were made from them.

The work was done in the Zoological Laboratory of the University of Kansas under the guidance of Dr. H. H. Lane. I wish to acknowledge my sense of obligation to Dr. Lane for his kindly interest and helpful suggestions, always willingly given.

DESCRIPTION.

To make clear the course of development of the mesonephros in Peromyscus leucopus, it seems necessary to begin the description with the period preceding the stage in which the nephrogenic tissue can be said to be actually mesonephric. The chief difficulty encountered lies in the fact that a debatable area occurs between those regions which are distinctly pronephric and mesonephric in character. There are, therefore, obvious advantages in the plan of first describing a stage in which the pronephros is present but in which the mesonephros has not yet begun to appear.

Embryo No. 40, G. L. 2.1 mm. (Fig. 1)

In embryo No. 40 the primary mesodermal structures are already fully formed. In the anterior half, approximately, of the trunk, the differentiation of the somites, nephrotomes, and lateral plates has occurred, but in the posterior portion differentiation is not so far advanced. The somites and nephrotomes have not been delimited, though the segmental plates are well defined, and are separated by the coelomic cavity into their somatic and splanchnic layers. The nephrotomic region, however, is indicated by a longitudinal groove

or depression in which the pronephric duct is developing. (Fig. 11)

A myocoel is not present in the fully formed somites. In each of these structures there is a central mass or core of irregularly arranged ovoid cells surrounded by an epithelium of radially arranged columnar elements. The nephrotomes have the form of a solid, unsegmented ridge on the dorsal wall of the coelom and does not inclose a portion of the coelom.

The coelom through a large portion of the trunk region is not closed below (Fig. 10). The splanchnic layer of the mesoderm is for the most part thin, being only one or two cell layers thick, and applies itself around the gut and aortae. The somatic layer is thick, has a somewhat stratified appearance, and applies itself rather closely against the ectoderm. The cells on the coelomic side of the lamina are fairly regular, tall and cylindrical. On the ectodermal side there is a gradual transition into mesenchymatous tissue. The somatic layer, leaving the body wall, makes a sharp angle inward and downward toward the dorsal aorta to the point where it joins the nephrotome and, as a result, it makes another sharp angle at the point of the junction with the nephrotome. It is in this angle that the pronephros develops (Fig. 10).

Toward the posterior end of the embryo the ventral wall of the body again closes and the coelom extends posteriorly as a pocket into this region on each side of the body. The arrangement of cells at the end of this pocket is the same as that of the somatic or splanchnic layers. In fig. 11 a section is shown through the posterior wall of the coelom on the left side. Behind this formed coelom and the body wall the space is filled with mesenchyme and in it the dorsal aortae follow around the ends of the coelomata to the ventral side. Shortly they unite to form one vessel which, after giving off a single umbilical artery, continues forward closely applied to the ventral side of the gut. Anteriorly this gives off three rather large branches which again empty into the left dorsal aorta. Shortly it leaves the body and continues its course until it ends in the vessels of the chorion.

The nephrotome, according to Felix (P. 754, Keibel and Mall, Human Embryology), presents quite different arrangements in the representatives of the various classes of vertebrates. Quoting him, "It may: 1, preserve the form of an epithelial canal and its connections with the lateral plate and be directly transformed into a uriniferous tubule; 2, it may become separated from the

lateral plate and form either an isolated vesicle or an isolated solid mass of cells, and in this form it may be directly transformed into a tubule; 3, it may be gradually taken up into the lateral plate by an extension of the coelomic cavity of the plate, and is then only distinguishable from the lateral plate by its further development; 4, it may be transformed into a solid mass of cells and fuse with the neighboring stalks, similarly transformed, to form a single cord, known as the nephrogenic cord, which may or may not retain its connection with the lateral plates; or 5, it may finally be transformed into mesenchyme tissue by the separation of its cells, this tissue then fusing with the mesenchyme of the sclerotome to form a single mass in which the source of the constituent parts can no longer be determined. Several of these five modes of development may occur in the same embryo, and, since neighboring segment stalks develop similarly, groups of segment stalks may be distinguished.

In Peromyscus leucopus embryos 3 2-3 mm greatest length the arrangement is most nearly like the fourth of the above groupings. There is no indication of segmentation in the nephrotome; it is joined to the somites and between the somites it continues as a solid ridge

of cells without any regularity in their arrangement, attached to the lateral plate (Fig. 10). There is no indication of a cleft or cavity within this ridge to indicate its origin from somatic and aplanchnic layers of mesoderm as described for the mole by Kerens. In the anterior end the cells become mesenchymatous and it is impossible to follow their fate.

Since the pronephric duct extends almost back to the cloaca, it is presumed that the pronephros is completely formed at this stage of development of the embryo. At the anterior end, in the glandular portion, it is dissolving into mesenchyme so that it is impossible to know how many tubules were formed. In one embryo, three, and in another four tubules may be counted. In two of the tubules the cells are arranged in a somewhat radial direction but there is no lumen; in all other tubules and in the duct the cells form a solid cord with no indication of a lumen.

The tubules appear as elevations which have their origins in the parietal angle at the base of the nephrotome and connect with the pronephric duct. The duct extends close under the ectoderm in the depression formed between the nephrotome and the somatic mesoderm of the inner wall of the coelom posteriorly almost the full length

of the trunk ending free near the posterior end of the coelom. It is still in close apposition to the ectoderm at its tip.

The posterior portion of the gut should be described briefly. Back of the anlage of the stomach it becomes rounded into a tube with relatively very thick walls of tall columnar cells. This tube continues backward to the posterior region of the trunk where it rapidly enlarges until its cavity is almost as large as the cavity of the coelom on either side. For a short distance it is in contact with the ectoderm forming the anal plate. Thence it continues posteriorly a short distance as the tail gut. There is no allantoic diverticulum nor any indication that there has been one. If a yolk sac has been present it has already been closed off so that in this embryo the gut is a continuous tube without evaginations outside of the body cavity.

Embryo No. 434, G. L. 3.1 mm. (Fig. 2)

In embryo No. 434, the first indication of a mesonephros is found. Remnants of the pronephros are still present. At its most anterior extremity the organ for a short distance is represented only by a solid, irregular but, for the most part, continuous cord in a disintegrating

condition. Then the remnants of two small, very degenerate tubules, one of them with a very small lumen, are attached to the cord and are followed for a short distance by the solid cord without any indication of tubules. Next four more tubules in succession are found. The first and second of these are in a disintegrating condition but each still shows a definite lumen in at least a part of the tubule. They are attached to the cord of cells (the pronephric duct) but the lumen does not extend into the duct. The duct is larger in diameter than it was more anteriorly but at this point is still a solid cord.

These six tubules are interpreted as the last two of the pronephros and the first four of the mesonephros, although it is impossible to say this with any certainty. The larger gap between the second and third tubule than between the third and fourth or the first and second may be considered as the boundary line between the pronephros and mesonephros and is apparently the intermediary zone described by various authors. It probably has some phylogenetic significance. Brauer describes this region as he found it in the Gymnophiona. In them it extends from the 15th to the 24th somites. Some rudimentary tubules develop in the region but never unite

with the Wolffian canal.

Janosik studied the development of the excretory organs in the spermophyle. The first rudiment of the organs consists of a thickening of the dorsal part of the intermediary piece beginning with the 6th somite. This thickening represents both the tubules and the Wolffian duct. It isolates itself progressively from the coelom becoming a cellular cord, which, on account of its position, remains attached to the lateral lamella by the ventral part of the intermediate piece, prolonging itself on a level with these unions in the coelomic groove. Soon all of the anterior part of this rudimentary organ atrophies, its cranial extremity not reaching more than to the 9th somite. Beginning with the 15th somite, the Wolffian duct grows free and elongates itself without uniting either with the mesoblast or the ectoblast. Janosik considers this rudimentary organ a pronephros. The region of the 12th to the 15th somite in which the cellular cord is no longer attached to the coelom is an intermediate zone. Finally in the 16th somite the mesonephros proper begins, the tubules issuing from the intermediate piece and connecting with the Wolffian duct.

The description of this stage given by Kerens for the mole corresponds in its general outline to the condition

found in embryos of Peromyscus leucopus and in the spermophyle. The intermediary zone is made up of a single nephrotome in the mole. In a further description by Kerens of the rabbit embryo, however, no mention is made of this intermediary zone.

Concerning this region of transition in the human embryo Felix makes the following statement, "The time of disappearance of the pronephros cannot be determined definitely, since it extends into the territory of the mesonephros, and the cranial tubules of these also degenerate immediately after their formation; it is consequently impossible to determine the significance of the remains of a tubule occurring in the pronephric region; it may represent a pronephric tubule, it may be a mesonephric tubule in process of degeneration, or, finally, it may be the remains of a primitive segment stalk or of the nephrogenic cord."

Felix thinks that the pronephros and mesonephros are entirely distinct organs derived from different parts of the mesoblast and that rudimentary mesonephric tubules may exist in the posterior region of the pronephros. However, according to Brauer these rudimentary mesonephric formations are the remains of the union of the nephrotome and the scleromyotome.

Nothing in the embryos of Peromyscus leucopus would indicate any overlapping of the pronephric and mesonephric regions.

The first and second tubules of the region which we will call mesonephric, as mentioned before, are in a disintegrating condition even before they have fully formed. The third and fourth go farther in their development. The diameter of the tubule in its widest part is greater than the diameter of the Wolffian duct. A large lumen has appeared in the central portion of the tubule which is composed of cuboidal epithelium. The tubule as a whole has a slightly hook-like shape with the part of greatest diameter at the bend of the hook. As will be described more in detail later, the tubule develops from a solid mass of cells which hollow out to form a vesicle. This vesicle then lengthens in two directions, partly by rearrangement of its cells and partly by growth. These two tubules, then, are seen to be in the very early stage of their formation. The cavity is extending into the projection toward the Wolffian duct but has not yet broken through into the cavity of that duct. It is only in the region of these two tubules that there is a cavity formed in the Wolffian duct at this time. Its anterior portion has already been described; posteriorly it continues as a solid cord, gradually reducing in size, in the same general position as in Embryo No. 40, with this difference that

it is no longer applied closely to the ectoderm due to the beginning of development of tissues, including the post cardinal vein, between it and the ectoderm. It ends freely with no closer approach to the cloaca than in embryo No. 40.

Along the duct posterior to the two tubules described above are four condensed masses of cells which are about to be transformed into vesicles. They are close to but not attached to the Wolffian duct.

Posterior to the cloaca a post-anal gut extends into the tail for a short distance. There is no indication of an allantois.

The development and differentiation of the mesodermal tissue as a whole is very rapid during the period represented by embryos No. 40 and No. 434. In No. 40 segmentation has progressed to a point five somites posterior to the last tubule of the pronephros. In No. 434 segmentation is completed throughout the full length of the body and well into the tail region. In the anterior region the somites have broken down into the myotome, sclerotome and mesenchyme. All traces of the intermediate piece are lost in this region making it impossible to know from what part of the nephrotome the mesonephric tubules are formed (Fig. 14). That this differentiation of

of the mesoderm is apparently much more rapid for Peromyscus leucopus than for the mole is indicated by the statement from Kerens in the description of embryos of 15 and 16 somites, that the last formed nephritic anlage appears at the same time as the last scleromyotome is formed.

Embryo No. 34. G. L. 4.2 mm. (Fig. 3.)

In the preceding embryo four anlagen of mesonephric tubules were found. In embryo No. 34 the early stages of eighteen tubules are present. No other organ shows such rapid development as the mesonephros during the interval between the stages represented by these two embryos. Segmentation has been completed. The somites have broken up into their myotome and scleretome portions. Due to the growth in thickness of the somatic mesoderm and the dorsal extension of the coelom, the mesonephric region and also the post cardinal vein appear to have migrated inward for a considerable distance (Compare figures 15 and 13.).

The large number of developing tubules in this embryo cannot be taken to indicate a wide gap in the developmental history between this and embryo No. 434. In that embryo segmentation was complete except for a

part of the tail region and most of the somites had been changed into their myotome and sclerotome derivatives and all indications of the intermediate piece were gone. The mesodermal tissue in this embryo, throughout most of the trunk region, is in about the same stage of development and we find the tubules throughout the length developing in such rapid succession that all appear to have originated at about the same time.

There is, however, a progressive development of the tubules, those in the anterior region being slightly more developed than those in the posterior region, although occasional tubules are found which are considerably more or considerably less developed than the ones just preceding and the ones just following them.

The first tubule in this embryo is almost completely disintegrated. The Wolffian duct has completely disappeared opposite it. No trace of the pronephric region is present. The tubules in their disintegration break down and their cells separate and mingle with the surrounding mesenchyme so that it is impossible to distinguish them from the mesenchyme.

The second tubule is also in a disintegrating

condition. It is a slightly elongated vesicle and its ragged cavity is continuous with the cavity of the Wolffian duct at the anterior tip of the duct.

The third, fifth, and sixth are the most highly developed of any of the tubules in this embryo. They have elongated and have become somewhat curved, almost sickle shaped. They have a well defined cavity which is continuous with the cavity of the Wolffian duct. The walls of the tubules are composed of well defined cuboidal epithelial cells.

The fourth tubule is smaller and shorter than either the third or the fifth. It has a slight cavity but not continuous with the cavity of the Wolffian duct. The tubule is joined to the wall of the duct however.

The seventh and eighth are in a stage of development about like the fifth with the exception that the cavity of the seventh is not connected with the cavity of the Wolffian duct while that of the eighth is so continuous.

The ninth and tenth are small and somewhat spherical with very slight cavities. In these not even the walls of the vesicles join the Wolffian duct. From the eleventh to the eighteenth the vesicles are more uniform, each one slightly less developed than the one which precedes it. All contain cavities but the cavities do not connect with the lumen of the Wolffian duct. Just posterior to

the eighteenth there is a small condensed mass of mesodermal tissue from which another vesicle is to be formed.

Fig.22 is a drawing of a reconstruction of the eleventh vesicle. It is rather flattened on its posterior surface and dome-shaped on the anterior surface. Posteriorly the succeeding vesicles appear to become gradually more spherical indicating that the first steps in the formation of a tubule from the spherical vesicle consists in a rearrangement of the cells of the vesicle.

Posterior to the last vesicle the Wolffian duct continues until it ends beside the cloaca. Near its distal end it enlarges so that its diameter is about three times as great as throughout all of the more anterior portion of its length. There is no indication of a metamorphic evagination at this time. Figs.12 and 13 show the relationship of the Wolffian duct to the cloaca. The duct ends blindly three sections (10 microns each) beyond the section from which this drawing was made.

The cloaca of this embryo is quite similar to that described for preceding embryos. There is no indication of an allantoic evagination from the hind-gut region. The post-anal gut shows a remarkable development. It now extends as a slender tube far into the tail.

Embryo No. 27. G. L. 5.2 mm. (Fig. 4.)

In the last described embryo (No. 34) the two most anterior tubules were found to be in a condition of disintegration even before they had more than just commenced to take on the form of a tubule. There was nothing anterior to these to indicate whether or not other tubules had existed.

In embryo No. 27 seventeen tubules or vesicles are present on the right side and eighteen on the left. Fig. 23 is a drawing of the reconstructed first tubule and end of the Wolffian duct on the right side. The duct begins abruptly, as a solid mass of cells staining more deeply than the surrounding cells in one section. In the next section is the open Wolffian duct with the wall of the tubule extending from it. There is no trace of nephritic remnants anterior to this point on the right side, but on the left, a solid, more or less continuous, but irregular cord of more deeply stained cells extends anterior to the first tubule for a short distance.

The first tubule is more developed and is larger than any of the tubules in embryo No. 34. The second tubule is similar as are also the third and fourth, with the exception that the cavities of these last two do not communicate with the Wolffian duct. The fifth tubule is

larger and longer. In form it has reached an S-shaped stage of development. It has a cavity throughout its length, which opens into the Wolffian duct. The sixth is only a slightly elongated vesicle entirely separate from the duct. The seventh and eighth are larger, being at about the same stage of development as the second tubule of embryo No. 34. Their cavities do not connect with the Wolffian duct. The ninth, tenth and eleventh are rather straight short tubes, entirely separate from the Wolffian duct. The twelfth to the seventeenth are similar in every way to the most posterior vesicles in embryo No. 34. Posterior to the seventeenth tubule a solid mass of mesodermal tissue is the first indication of still another tubule.

The Wolffian duct continues posteriorly, enlarges in diameter, and ends beside the cloaca without opening into it in the same manner as described for embryo No. 34. In this embryo, however, the fundamentals of the metanephros are present. A slight evagination from the dorsal wall of the Wolffian duct is the beginning of the future ureter. A condensed mass of more deeply stained cells from the nephrotome form a cap over this evaginated portion.

Embryo No. 435. G. L. 6.1 mm. (Fig. 5.)

In embryo No. 435 the entire mesonephros has enlarged so that the mass protrudes into the coelom and all of the tubules have lengthened. The epithelial layer on the ventral side of the mesonephros which is the anlage of the gonad is quite apparent but is still a more or less flattened layer with no indication of a constriction to separate it from the mesonephros.

The majority of the tubules now are more nearly in the form of the fully developed tubule. Mihalkovics (loc. cit.) describes this shape for the mesonephros tubule of the sheep as a double or reversed spiral. In his description of the development of the tubules in the sheep he states that the tubule attains an S-shape and then a period of growth in the region of the middle bar of the S results in the recurved spiral.

In the tubules of the mesonephros of Peromyscus leucopus it is impossible to say just how the final form is attained. Some tubules are found which have the S-shape without having as yet any indication of the recurving of the middle bar. In others what is to be the middle bar of the S is growing up to form the recurved portion before the lower bar of the S is formed, that is, their form is that of a simple spiral.

In this embryo (No. 435) the anterior portion of the Wolffian duct has disappeared. Anterior to the end of the duct are the remains of one tubule in the form of a compressed hook lying at about right angles to the direction of the Wolffian duct. The next six tubules are in variable stages of degeneration but they can all be followed throughout most of their length. None are attached to the Wolffian duct. In this anterior region the tubules appear to have rearranged themselves or to have been twisted with respect to the long axis of the mesonephros, so that now, a cross section through the embryo cuts the tubules at several levels. In the preceding embryos described and in the more posterior part of this embryo the cross section through the embryo usually cuts through a considerable length of the tubule. It is as though this anterior portion of the mesonephros had been stretched, pulling the tubules in such a way that they no longer lie in a plane at right angles to the length of the organ.

The tubules from the seventh to the twelfth are all similar in their general development although some are slightly longer than others. All are connected with the Wolffian duct and their lumina open into its cavity. The following four tubules, the twelfth to the fifteenth,

also are quite similar to these with the exception that they are not quite connected with the Wolffian duct. From the sixteenth on to the last tubule, the twentieth, all of the tubules are connected with the Wolffian duct. All are fully developed with the exception of the last which is short and slightly bent, not yet having attained even an S-shape. It does, however, have a cavity throughout its length which opens into the lumen of the Wolffian duct.

Behind the last tubule the Wolffian duct continues posteriorly ending as a blind tube beside the cloaca without opening into it as described for the preceding embryos, with the exception that, in this embryo it is not enlarged in its distal portion, but has about the same diameter throughout its entire length.

The metanephric anlage shows considerable development. It now consists of a rather long slender ureteric portion which at its distal end enlarges into a sausage-shaped vesicle which is at approximately right angles to the ureteric portion. One end of this enlarged vesicle shows the first indication of forking. A condensed, deeper staining mass of cells, the cortical anlage, forms a cap over this vesicular portion. Fig. 19 is through the length of the ureter and shows the cross section of the vesicular portion.

Embryo No. 42. G. L. 6.4 mm. (Fig. 6.)

In this embryo a greater number of mesonephric tubules, (24), were found than in any other stage. It cannot be said that the mesonephros has reached its maximum development at this time since, as will be seen in the description of the next stage, greater development is attained especially in the posterior portion of the mesonephros.

It is also impossible to say with certainty that twenty-four is the maximum number of tubules developed because, as has been mentioned previously, degeneration at the anterior end is so complete that all traces of the pronephros disappear very early; likewise the Wolffian duct completely disappears in some cases farther posterior than the remnants of mesonephric tubules which have presumably been connected with it before their degeneration. Accordingly it is possible to record only the number of tubules or remnants of tubules found in each stage. For those degenerating tubules at the anterior end there are two possibilities, either they completely disappear or some of them may persist and enlarge with the growth of the entire gland, even though they have separated from the Wolffian duct and are gradually disintegrating.

In this embryo the first seven tubules are all in a state of disintegration. Only fragments of the first tubule persists with no indication of its connection with the Wolffian duct. In the seventh, the connection with the Wolffian duct still persists. The eighth tubule is connected with the Wolffian duct as are also the ninth and tenth. These three all have the typical S or the recurved spiral shape.

In the eleventh and twelfth, we find the first definite indication of what can probably be interpreted as the formation of secondary tubules. Neither the eleventh nor the twelfth connect with the Wolffian duct. The proximal end of the twelfth is just beneath that of the eleventh. The eleventh tubule has the shape of a simple spiral, while the twelfth is that of the recurved spiral and lies entirely between the loop of the eleventh and the outer wall of the mesonephros and in the same plane, so that both tubules are present in the same sections.

The thirteenth tubule joins the Wolffian duct while the fourteenth does not; the fifteenth joins but the sixteenth, which is just ventral to the fifteenth, similar to the condition found in the eleventh and twelfth, does not join. The seventeenth and eighteenth also come off from the Wolffian duct at the same point, the eighteenth

just ventral to the seventeenth. The eighteenth has the form of the simple spiral.

The nineteenth and twentieth both have the form of the reversed spiral. Neither are connected with the Wolffian duct. The twenty-second is just ventral to the twenty-first, both joining the Wolffian duct. This tubule is of special interest. It is presumably a secondary tubule and has the form of the reversed spiral. At the distal end, however, differing from all other tubules previously described, this tubule terminates in a Bowman's capsule. One tubule, opposite this one in the right mesonephros, also ends in a capsule; however, the latter is not so well developed. The tubule narrows down suddenly to form a short neck then expands into the capsule. One of these capsules from another embryo is shown in Fig. 20. The inner wall is thick, composed of large, deeply staining epithelial cells with oval nuclei. At least a portion of the outer wall of the capsule is composed of greatly flattened lightly staining cells. A capillary enters the capsule and enlarges filling the cavity. In the case of this one tubule the capillary is possibly arterial. Just opposite the tubule a capillary is given off from the dorsal aorta and starts its course in a direction in line with the capillary from the capsule but there is a space between the

two which it is impossible to follow. This case demands special mention because in all other Bowman's capsules found, the capillary is unquestionably venous.

The last two tubules of the organ, the twenty-third and the twenty-fourth do not join the Wolffian duct. The twenty-third has the reversed spiral form but the twenty-fourth is small and in a very early stage of development.

Concerning the blood supply of the mesonephros, it should be noted that in all embryos up to this stage and with the exception of three capillaries in this one, there is apparently no arterial blood supply. The posterior cardinal vein communicates at frequent intervals with the subcardinal. It is however impossible to follow this vein in any embryo. It appears through a few sections at the points of communication with the posterior cardinal and then disappears again. It is impossible in these embryos to follow any of the small capillaries unless they contain blood cells, but one would not expect to find this same condition in a vein as large as the subcardinal at the point of communication with the posterior cardinal (Fig. 15).

The aorta, as mentioned, does not give off any branches to the mesonephros in the earlier stages. In the embryo under discussion one capillary enters the mesonephros on the right side opposite the seventeenth tubule. Opposite

the twenty-second tubule on the left side the capillary already described is given off and one more on the right side opposite the twenty-fourth tubule enters the mesonephros.

The differentiation of the cloaca into the rectum and urogenital sinus is well under way in this embryo. A description of its development, however, throughout the entire series of embryos will be given later.

The gonads are now rounded thickened masses of deeply staining cells but without any indication of sexual differentiation. A slight indentation or groove on the inner side has commenced to mark off the gonad from the Wolffian body.

Embryo No. 280. G. L. 6.5 mm. (Fig. 7.)

In embryo No. 280 the urogenital fold differs very little in its general appearance from the condition found in the last preceding stage. Sexual differentiation has not yet occurred, the gonad being only slightly larger than in embryo No. 42 and not yet as large in cross section as the mesonephros. (Fig. 16.)

The first six tubules have degenerated but all have their proximal ends still attached to the Wolffian duct. In most instances observed the connection with the Wolffian

duct is lost almost as soon as degeneration sets in. The first tubule in this embryo is a short, only slightly curved tube, but the others have all developed to a re-curved spiral form.

The tubules from the seventh to the tenth are all well formed and connect with the Wolffian duct. Fig. 24 is from a reconstruction of the ninth tubule. The eleventh has formed Bowman's capsule; there are, however, no capillaries associated with this capsule.

From the twelfth tubule on to the last, the twenty-third, all have the suggestion, at least, of a Bowman's capsule with the exception of the fourteenth. There are no capillaries associated with the number of these capsules however. The eighteenth tubule is of interest because it branches, the branching occurring at the beginning of the first curve from the Wolffian duct. This is the only branching tubule observed during the entire study.

It should be pointed out here that the Bowman's capsule as described is not to be confused with the typical picture which one has of this organ. Here, at best, it is a simple cup as illustrated in Fig. 20, and with one exception mentioned in the description of embryo No. 22 none more developed was observed in this or older embryos. In the majority of cases development has not gone so far,

that is, the cup is not so well formed. In most cases it is necessary to examine the tubule carefully to detect the capsule. If at the end of the tubule the outer wall was composed of flattened cells and the inner of those of the rather cuboidal type, a capsule was recorded.

In Peromyscus leucopus we apparently find a condition slightly in advance of the rat. Weber has described the mouse, (Mus musculus) as not having any indications of glomeruli in the mesonephros. Bremer briefly mentions that in the rat the tubules may end in a blind enlargement while here, in Peromyscus leucopus we find in the posterior end of the mesonephros, just before that portion begins its disintegration, that Bowman's capsules are appearing even though they are in the most elementary stage of development. One other point of interest in connection with these capsules is the fact that here they are forming frequently without any blood capillaries in contact. In all descriptions of development of the Bowman's capsule the arterial capillary in contact with the end of the tubules is given as the causative factor or stimulus in the shaping of the Bowman's capsule. Another point of interest is the fact that in all cases in Peromyscus leucopus, with one possible exception, if a capillary was in contact with the capsule that capillary was venous.

The Wolffian duct in No. 280 does not yet open into the urogenital sinus. This is rather remarkable since in the embryos of other species of mammals described, the duct enters the sinus relatively early, usually by the time the sinus has begun its differentiation.

The metanephros is developing rapidly. Several of the collecting tubules are forming and branching in the more or less typical T form and have condensed masses of cells beneath the outer ends of the arms of the T. However, no uriniferous tubules have as yet started to form.

Embryo No. 21. G. L. 8 mm. (Fig. 8.)

In the last preceding embryo (No. 280) the mesonephros apparently has reached its maximum development. In No. 21 there is little to record that is different. The gonad is slightly larger but sexual differentiation cannot be determined as yet (Fig. 17).

The first three degenerate tubules appear anterior to the end of the Wolffian duct. The fourth, also degenerate, is opposite the end of the Wolffian duct. The fifth to the ninth do not join the Wolffian duct. The tenth and eleventh do join the Wolffian duct but are degenerating, while the twelfth does not connect.

All tubules from the twelfth to the twentieth are connected with the Wolffian duct although the nineteenth is a very incompletely formed tubule. The twentieth is still in the stage of a small vesicle.

The structures which have been described as capsules, even though they are so immature as to be hardly recognizable, are found on the distal end of all tubules from the ninth to the eighteenth. Four of them have no sign of capillaries in them.

The Wolffian duct in this embryo opens into the urogenital sinus. As will be mentioned in connection with the development of the urogenital sinus this opening is very late when compared with the condition in other mammals.

The metanephros has not made any noteworthy changes beyond the condition of development described for the preceding embryo. The collecting tubules are developing and becoming more numerous but none of the uriniferous tubules can be found.

Embryo No. 22A. G. L. $11\frac{1}{2}$ mm. (Fig. 9.)

The study for this stage is based on two embryos from the same uterus, one a male and the other a female. Unless mentioned, the description is from the male embryo.

The mesonephros is degenerating throughout its entire length while the gonads and the Müllerian ducts are in a condition of growth and differentiation. The mesonephros has undergone greater degeneration in its middle than in the anterior or posterior portions so that only occasionally the faintest traces of fragments of tubules are to be seen in this portion. At both ends of the organ the tubules are all fragmented but are not so completely disintegrated. In the male embryo, at the anterior end, the first four tubules are connected with the Wolffian duct. They cannot be followed beyond the point of the beginning of the first curve although other sections, presumably of the same tubules, are present. Some of the sections of these tubules lie just dorsal to a deeper staining mass of cells, the anlage of the rete testis, which separates the testis from the mesonephros (Fig. 18). No continuity between the tubules and the anlage of the rete testis has been established at this stage of development. In the female embryo none of the tubules in this anterior region are connected to the Wolffian duct. The anlage of the rete ovarii is present but is not so prominent as its homologue in the male embryo.

In the posterior region of the mesonephros, the tubules are all fragmented so that it is impossible to follow any one of them. A point of considerable interest in connection with these posterior tubules is the development of a few glomeruli into a more mature condition than found elsewhere throughout the entire series. In the male embryo four, and in the female two, glomeruli have continued to develop until they have almost the appearance of the typical glomerulus. Developing glomeruli in the metanephros, which are to be seen in the same section, are quite similar in appearance but are about three times as large. It seems probable that some of the glomeruli in the mesonephros may continue their development even after separation of the tubule from the Wolffian duct and even after fragmentation of the tubule has begun. All other glomeruli in the mesonephros of these two embryos have completely disappeared.

The Wolffian ducts and the ureters have become separated by the absorption of their ends into the walls of the urogenital sinus and now open into the urogenital sinus at points separated by a distance of fourteen sections of 10 microns each. (Figs. 29 and 30.) The Müllerian ducts which will be described presently have not yet reached the urogenital sinus.

As mentioned above, the gonads have been undergoing rapid growth and differentiation. The testis in the male embryo is slightly shorter but broader than the ovary in the female embryo. In the testis the testis cords are quite apparent (Fig.18). In the ovary the primordial germ cells are scattered promiscuously throughout the organ and around them are many circles of epithelial cells indicating the beginning of the development of follicles.

The metanephros is also developing rapidly. The uriniferous tubules are becoming convoluted and glomeruli are becoming prominent in the cortical region. One would not, however, consider it possible for the metanephros to function as an excretory organ at this stage of its development. Certainly, if such an assumption can be made on a purely histological study, the mesonephros has never attained a stage of development where it was functional for excretory purposes. We are led then to consider that up to this stage, at least, excretion must be accomplished by some other organ and we agree with Bremer (1916) that the placenta appears to be the most probable organ for this function.

The Müllerian duct is first met with in these embryos. Fig.21. is the ninth section (10 microns thick) through

the urogenital fold and shows the funnel-shaped pit which forms the ostium of the Müllerian duct. The ostium tubae abdominalis in the female embryo is so similar as to be practically identical with that of the male. In the male the pit extends through the first eight sections; in the female through seven sections. One point of interest concerning the ostium tubae is that it opens on the ventral side of the urogenital fold. Prentiss and Arey (Fig. 222, P 211,) show a section through this region for a human embryo of 10mm. length and Kiebel and Mall (Fig. 622, P. 912,) show a similar section through a human embryo of 11 mm. length. In both illustrations the groove is on the dorsal or dorso-lateral side of the fold.

The Müllerian duct continues posteriorly in the angle at the outer margin of the mesonephros, lateral to the Wolffian duct. Near its posterior end it crosses over the Wolffian duct and comes to lie mesad to this duct. Fig. 28 shows the duct in the urogenital fold at the region where it is crossing over the Wolffian duct. The points marked X are the ends of the ducts in this embryo. Concerning the method of growth of the ducts nothing could be determined from these embryos. Felix (Keibel and Mall P. 913) describes the process of growth

of this duct for the human embryo and since it is very probable that the process is the same in this mouse we will quote him:

"At the very time when the posterior end of the groove is separating from the epithelium it begins to grow out caudally and in this process we come to the development of the distal portion of the Müllerian duct; it is formed by the gradual outgrowth of the tip of the cornet. The path that it will follow is already laid out; it must pass, caudally between the coelomic epithelium laterally and the primary excretory duct medially, as if between two bars. Its growth results entirely from its own forces, the outgrowing tip being always free and connected neither with the coelomic epithelium nor the primary excretory duct; only poorly preserved or poorly fixed preparations could deceive one on this point. The growth depends on the increase of cells partly along the entire extent of the duct as is shown by the mitoses, partly at the outgrowing end, which is frequently swollen and presents all the peculiarities of a so-called cone of growth,----- The lumen extends continuously from the funnel into the caudal portion of the duct, and follows the growth of the duct so closely that all that is ever solid is a small terminal portion."

The Urogenital Sinus.

The development of the urogenital sinus in the deer mouse, (*Peromyscus leucopus*) is of interest because of the fact that from the hind gut no allantois is ever developed. In order to make the description of the developmental stages more coherent these structures were reserved for this separate section.

In embryo No.40, the neural tube extends to the tip of the tail although it is an open medullary plate in the last ten sections. Sixteen sections from the tip of the tail the posterior end of the gut appears abruptly in the cross sections. The gut has enlarged greatly in the posterior part of the body. It is flattened laterally but extends from the notochord above to the body wall below as seen in cross sections. (Compare Figs.10 and 11).

The Wolffian ducts have not reached the cloacal walls.

In embryo No.434 the general conditions are the same except for those caused by the growth and curling of the embryo. At the posterior end, the gut, instead of ending abruptly, reduces in size and extends out into the tail for a short distance as the post-anal or tail gut.

In embryo No.34 the curling of the body has continued to the extent that all of the enlarged posterior

portion of the gut lies ventral to the smaller intestine in all sections through this region. The tail gut is a long slender tube extending through fiftyfour sections of ten microns each. The Wolffian duct now lies close to but does not open into the cloaca (Fig.12). The point at which it first comes in contact with the wall of the cloaca differs somewhat in the embryos of Peromyscus leucopus from that in the human embryo. Felix mentions that in the human embryo the first contact is on the side of the cloaca near the body ectoderm and that later the end of the duct moves upward to a point about midway on the cloaca as seen in cross section. In Peromyscus leucopus its point of first contact is in this middle region (Fig.13).

Embryo No.27 shows no changes with the exception that the tail gut is now shortened to one third of the length which it had attained in embryo No.34.

The ventral wall of the cloaca has come in contact with the ectoderm for a few sections forming the beginning of the cloacal membrane. As growth continues this membrane becomes more extensive.

Between embryos No.27 and No.435 the beginning of the separation of the urogenital sinus occurs. In embryo No.435 the tail gut has completely disappeared

except for just enough of a projection to serve as a landmark. The initial separation of the cloaca into rectum and urogenital sinus in the human embryo has been described briefly by Prentiss and Arey (Text book of Embryology, page 206) as follows: "The saddle-like partition, between the intestine and the allantois grows caudally, dividing the cloaca into a dorsal rectum and ventral, primitive urogenital sinus."

In Peromyscus leucopus, as previously mentioned, there is no allantoic evagination from the hind gut. Nevertheless, the urorectal septum enters like a wedge of tissue forced into the ventral side of the cloaca and directed toward the posterior end; it passes just dorsad to the points where the ends of the Wolffian ducts lie.

While this separation of the urogenital sinus from the rectal portion takes place a region of rapid growth has appeared on the sides of the portion of the urogenital sinus below the points where the ends of the Wolffian ducts lie against the urogenital sinus. There is no growth in the portion above the ends of the Wolffian ducts. As a result this part of the side wall on each side is carried up above the roof of the urogenital sinus as a pocket-like evagination. As a result

of the absence of growth in the portion of the side wall above the ends of the Wolffian ducts, this portion is shoved up and over so that it now forms the inner or medial wall of this evagination. The ends of the Wolffian ducts are carried along in this process so that they now lie on the mesial sides of these evaginations. Figs. 25 and 26 are both from the penultimate section of the Wolffian duct in each embryo and show clearly this relationship of the end of the Wolffian duct to the evaginated portion of the urogenital sinus.

The evagination points posteriorly, sloping gradually down to the urogenital sinus anteriorly in embryo No.435.

In embryo No.42 the evagination of the lateral wall has continued anteriorly until now it is almost as long as the urogenital sinus itself. There is no other feature of note in this embryo or the next of the series, No.280, except the growth in length of the urogenital membrane. The membrane now occupies almost the entire length of the urogenital sinus.

In embryo No.21, the growth from the sides of the urogenital sinus has stopped but the roof, which has been the bottom of the groove between the two evaginations, has grown or been raised so that a section

through this region no longer shows two evaginations from the sides but rather one large cavity. This elevation of the roof has again moved the Wolffian ducts so that now they are just behind the evaginated portion. They have at last fused with the walls and their cavities open into the cavity of this evaginated portion on its posterior wall (Fig.27).

This entire evaginated portion now begins to grow rapidly while the basal portion from which it originated remains practically at a standstill. The growing portion terminates anteriorly just posterior to the region where the umbilicus leaves the body in embryo No.22.

The terminal end of the Wolffian duct has been carried forward by this growth and has been absorbed into the walls so that now the ureter and the Wolffian duct enter separately (Fig.29 and 30). The Müllerian duct has not yet reached the urogenital sinus (Fig.28).

The urorectal septum has continued its growth until the rectum and the urogenital sinus are connected only by a small duct, the urogenital duct.

Summary

So far as known, there are only two groups of vertebrates, and only certain species of these, in which the mesonephros develops without the formation of glomeruli in connection with its tubules. One of these groups is found in the anamnia, the other in the amniota. In the first group are a limited number of teleost fishes; in the second are a few species of rodents.

The teleost fishes may be divided into three classes according to the glomerular development in the kidney.

In the first class, there are certain species which have no glomeruli in the mesonephros.

In the second class, mesonephric tubules are developed, some of which are connected with glomeruli, while others are not. Within this class there is a wide variation. Some species have only a very few glomeruli in the kidneys while in other species the glomerular tubules predominate.

The third class consists of those teleosts in which the kidney is entirely glomerular. This class includes the majority of the bony fishes. It is, however, of interest to note that in their embryo-

genesis, the mesonephroi of these fishes are entirely aglomerular (Audige fide Edwards) despite the fact that their promephroi are always glomerular. The glomeruli begin to make their appearance, depending on the genus, family or suborder of fish, according to Edwards (loc.cit). at about the time of the onset of sexual maturity.

The mesonephros is the functional kidney throughout the adult life in the teleost fishes. Careful comparative study of these fishes has shown that apparently the aglomerular kidney functions as effectively as the glomerular of the partly glomerular kidney. Cytological studies of the tubules of these kidneys show as great regional differentiation in cellular structure and just as certain indications of cellular activity in the tubules of the aglomerular as in the glomerular kidneys.

Other studies on fishes (Smith, loc. cit.) have shown that, even in fishes with glomerular mesonephroi, other organs, in this case the gills, may play an important role in removing from the body those metabolic products which in most vertebrates are eliminated only through the activities of the kidneys.

Among most of the amniot²as the mesonephros, if functional at all, serves as a kidney during a part only of the embryonic life of the animal. Thus it

has been rather definitely established that the mesonephros functions as a kidney in the reptiles and birds. In some of the reptiles the organ persists and functions for a considerable period after hatching, until the metanephros becomes sufficiently developed to take over the activity. In the opossum, among the mammals, the mesonephros continues to serve as the functional kidney after birth but while the young are still in the brood pouch. In the pig, the mesonephros reaches a greater degree of development than in any other mammal which has been examined and all histological studies indicate that it serves as the excretory organ during embryonic life. It has been pointed out previously, however, that there are a number of mammals in which it is very doubtful if the mesonephros ever serves as an excretory organ and some others in which it is unquestionably nonfunctional. Peromyscus leucopus is in this last group.

The criteria for assuming that the mesonephros is, or is not, functional during embryonic life in mammals depend entirely on morphological evidence.

Weber (loc. cit.) assumed, since the mesonephros in most of the forms which he studied degenerated before the metanephros was sufficiently developed to serve as

an excretory organ, thus leaving a gap where there was no functional kidney, that the mesonephros was nonfunctional in mammals, reasoning that if it were functional at one time it would continue to function until the metanephros was ready to take over the work. The pig he considered as an exception which he did not attempt to explain.

For his criteria of functional activity Bremer (loc. cit.) points out the correlation between the size of the mesonephros and the time of its degeneration, the amount of development of the allantois, and the development of certain cell plates in association with the blood vessels of the placenta which he terms a placental excretory organ. In determining the amount of development of the mesonephros he took special notice of the number and the size of the glomeruli.

Using this criterion he has arranged in a series the mammals which he studied. At the top of the series he places the pig in which the activity of the mesonephros overlaps that of the metanephros. The sheep also has a mesonephros which appears capable of functioning until after the metanephros is developed. In the rabbit and man excretion is probably carried on through the mesonephros in the early embryonic life but later through

the placenta. In the guinea pig during the early period of development the mesonephros may possibly serve as an excretory organ but Bremer doubts this because of the small size and fewness of the glomeruli. The mesonephros of the rat and the mouse, (using Weber's description of the mesonephros of the mouse) he decides, never serves as an excretory organ.

In Peromyscus leucopus then we apparently have a condition intermediate between that of the rat and the guinea pig.

- This paper has dealt with the development of the mesonephros in the deer-mouse with the purpose of finding out if it ever attains a development sufficient to permit it to serve as an excretory organ during any part of the embryonic life.

Differentiation of the Mesoderm

The mesoderm is differentiated and the somites are formed in Peromyscus leucopus in a manner quite similar to that of other mammals which have been described, with the possible exception that in Peromyscus leucopus the process appears to be more precocious. Splitting of the lateral plate into the somatic and splanchnic layers occurs before there is any indication of somite formation.

The somites are split out of the mesodermal plates as in other animals but no myocoel is formed. No indication of a metameric arrangement of the intermediate cell-mass, or nephrotome, could be found. The cavity of the coelom does not extend into the intermediate cell-mass which is a solid ridge of irregularly arranged, polyhedral cells. Almost at once, after their formation, the somites separate themselves from the nephrotome and are resolved into the myotome and the mesenchymatous sclerotome. The intermediate cell-mass also differentiates into mesenchyme immediately.

The Pronephros

The pronephros is found to develop as a mere rudiment: three or four solid projections, the vestiges of pronephric tubules, appear in the angle between the dorsal wall of the coelom and the nephrotome, apparently with their origin in the nephrotome. These projections bend and grow posteriorly, each uniting with the next posterior one. The free terminal end of the last one extends itself posteriorly just beneath the ectoderm as the rudiment of the pronephric duct. It does not reach the cloaca. This pronephric rudiment is very temporary and all traces of the

anterior or so-called glandular portion have disappeared before the mesoderm has differentiated into somites, except in the most anterior region. The vestige of the pronephric duct which is at this stage a solid cord remains and continues to develop into a tube which will become the mesonephric (or Wolffian) duct.

The Mesonephros

The mesonephros is formed along the dorso-lateral wall of the coelom. While the tubules are small, it has sufficient space in the retroperitoneum. Soon, however, the tubules enlarge and lengthen and the amount of mesoderm between the tubules increases to such an extent that it is forced to obtain more room. This it does by protruding into the coelom and forming a ridge or fold, the urogenital fold, in the dorsal body cavity. In its later development the urogenital fold contains the reproductive gland and the Müllerian duct as well as the mesonephros and its duct.

Somite formation has been completed throughout the length of the body and the somites have broken down in the anterior portion before the first of the mesonephric tubules begin to form. Throughout the entire length, the breaking down of the somites and

the intermediate cell-mass occurs before the mesonephric tubules are formed, making it impossible to tell from what portion of the nephrotome the tubules originate.

The post cardinal vein makes its appearance between the body ectoderm and the excretory duct at about the time the first mesonephric tubules begin to develop.

The first indications of the mesonephric tubules appear rather suddenly and almost simultaneously throughout most of the trunk region developing out of the mesoderm which previously formed the nephrotome. That there is a progressive appearance and development of these tubules, beginning at the anterior end and continuing posteriorly, is indicated by the slight advance in development of the anterior tubules over the more posterior ones. In Peromyscus leucopus embryos, however, this difference is very slight. In other forms of mammals, according to the published accounts, the tubules are formed immediately after differentiation of the mesoderm has occurred and it is possible in some, the mole for example, to find tubules in all stages of development in one embryo.

Degeneration of the more anterior tubules in Peromyscus leucopus takes place almost as soon as they are formed and before there is any indication

of the mature pattern. In all of the embryos studied some of the anterior tubules were degenerating, yet other tubules continue to appear posteriorly as the embryo grows older. The disintegration of the tubules at the anterior end is complete and not a trace of the tubules or of the Wolffian duct is left. This makes it impossible to know how many tubules have disappeared. Embryo No.42, (G.L. 6 mm) contained twentyfour tubules which was the maximum number found in any one embryo.

The Tubules of the Mesonephros

A mesonephric tubule originates as a solid mass of mesodermal cells which take a deeper stain than the surrounding tissue. The mass becomes hollowed out and at the same time becomes somewhat flattened on its anterior surface. By growth, but also largely by a rearrangement of the cells, projections are formed from this mass. One extends toward the Wolffian duct, the other away from it. The initial form then, as it becomes a tubule, is that of a "tilde", or wave, with the central portion thicker than either end. When contact is made with the Wolffian duct, the cavity of the developing tubule breaks through and communicates with the lumen of the duct. It seems

probable that some of the tubules fail to connect with the Wolffian duct, since frequently in the middle region of the mesonephros, more or less distorted tubules are found which are not joined to the duct, while other tubules both anterior and posterior to these are connected with the duct.

The distal end of the tubule continues to grow until in its most frequent mature form the tubule attains the shape of a reversed spiral. There is some question as to just how this final pattern is attained. In the sheep, according to Mihalkovics (loc.cit.), the tubule develops first to an S-shape and then a second S, in a plane horizontal to the first, develops in the middle bar of the original S resulting in the double S or reversed spiral form. In Peromyscus leucopus some tubules were found in the younger embryos which had an S-shape, but others had the form of a simple spiral of a little more than one turn. Both types were still developing, so that it would seem probable that in some cases the recurved spiral was derived from the S as described for the sheep, while in other cases the tubule developed directly into the recurved pattern.

The Glomeruli

Shortly after the tubule is formed from the vesicle and while it is in the early stage of its development, a Bowman's capsule and glomerulus begins to form at the distal end of the mesonephric tubule in all mammals which have been examined, with the exception of the mouse and the rat.

In *Peromyscus leucopus* no capsules are ever found on the tubules in the anterior half or two-thirds of the mesonephros. In the posterior region rudimentary capsules are formed on a part of the tubules during the last stages of development of the organ and after the tubules have acquired their mature pattern.

The tubule usually constricts, forming a short neck, and then expands into the capsule. The capsule is cup-shaped. The cells of the outer wall are greatly flattened and take a very light stain; those of the inner wall are large and cuboidal, with oval nuclei and stain deeply. These capsules were found for the first time in embryo No.42 (G.L. 6.4 mm). In this embryo one tubule, the third from the last, in each mesonephros ends in a Bowman's capsule. A capillary enters each of these capsules and expands filling the cup-shaped cavity. It is possible that this capillary

is arterial. As mentioned in the description of this embryo (P.58) an arteriole is given off from the aorta just opposite this tubule and starts its course in a direction in line with the capillary from the capsule. Just before the arteriole reaches the margin of the mesonephros, however, it disappears and cannot be followed farther. The capillary from the capsule also disappears before it reaches the margin of the meso-mesonephros. Thus there is a short space between the two where it is impossible to follow either of them or to determine whether they are both parts of the same vessel.

If this one capsule is supplied with arterial blood it is an exceptional case, because, in all other capsules observed in this study, the blood supply is unquestionably venous.

In the next two stages, embryos No.280 and No.21 (G.L. 6.5 mm and 5 mm respectively) the majority of the tubules in the posterior region of the mesonephros have Bowman's capsules. In many instances careful observation is necessary to detect these capsules because in the early stage of their development the diameter of the capsule is only slightly greater than that of the tubule and also because there is little

differentiation between the cells of the outer and the inner walls of the cup at the beginning of the formation of a capsule.

A number of these capsules were developing without any blood capillaries in contact with them. In the other capsules, a capillary derived from the post cardinal vein enters the cup and usually expands to fill the cavity, but does not form a glomerulus.

In no case, with the one exception mentioned above, is there an arterial capillary given off from the aorta even into the general region of the mesonephros where the Bowman's capsules are developing and in no embryo examined were there more than three very small capillaries from the aorta which even might enter the mesonephros. These could not be followed for more than a short distance, but in the embryos in which they were found, the suprarenal glands were developing rapidly and owing to their location, it is entirely possible that the capillaries were supplying them and were not entering the mesonephros.

Two facts just mentioned are of special interest. First, in the usual descriptions of the development of Bowman's capsule, an arterial capillary in contact with the end of the developing renal tubule is given as the

causative factor in the shaping of the capsule. In connection with the development of the general tubule pattern in the tubules of the kidney, no explanation is ever attempted as to why the tubule takes the form of an S or of a recurved spiral or the further complications of these patterns as found in the tubules of the mesonephros of the pig. Nevertheless with Bowman's capsule, presumably because in all previous observations in which its development has been described, it has developed in contact with a capillary network or glomerulus, it has been customary to consider the contact between the end of the tubule and the glomerulus as the cause of the invagination of the capsule.

Those tubules, then, in the embryos of Peromyscus leucopus which develop a Bowman's capsule without a capillary in contact with them, even though the capsule is very rudimentary, show conclusively that the formation of the capsule at the end of a renal tubule is a part of the general plan or pattern of development of the tubule and that the mechanical effect of pressure of the blood capillary is not essential to invagination.

The second point of special interest is the fact that the vascularization of the mesonephros in the

embryos of Peromyscus leucopus is entirely venous. It is probable that a similar condition or at least a condition of very limited arterial blood supply exists in those other rodents which have a non-functional mesonephros, however, the authors describing them have failed to discuss the source of the blood supply. In so far as the writer has been able to find, the only other instance which has been described in which glomeruli have developed in a mesonephros supplied with venous blood only is that of the salmon described by Haller (1908). Edwards (1928A loc.cit.), however, seems to question the accuracy of Haller's observations as is seen from the following paragraph. "The presence of arterial capillaries apparently involves the development of at least a few glomeruli (Lophius). The writer, therefore, shares the opinion of Audige and Guitel, which is somewhat in opposition to Haller ('08) that, in order for glomeruli to be present in the kidney, there must be arterial vascularization. Haller thought that he observed glomeruli which were developed from venous capillaries in the teleost, Salmo irrideus."

In the stage of development of Peromyscus leucopus embryos under discussion, to be sure, no true glomeruli are present, however, the fact that a capillary enters

the cavity of the capsule in most instances where a capsule is formed leads us to believe that if development were to proceed farther a typical glomerulus would be formed. In fact, as previously mentioned, in embryo No. 22, the last embryo of this series, three or four more nearly typical, but immature Malpighian bodies were found associated with parts of tubules which had separated from the Wolffian duct and were degenerating.

These Malpighian bodies are much smaller than some others which were developing in the metanephros as seen in the same section, however, each contains a glomerulus. It is impossible to follow the capillaries in the sections and for that reason, it cannot be said with certainty whether the glomeruli are of venous or of arterial origin. The fact that in all cases in younger embryos where the vascularization could be determined it was venous possibly makes it permissible to assume that these glomeruli are also venous.

At any rate, basing our judgment on these studies, we are led to agree with Haller that glomeruli may be formed without arterial vascularization.

The Malpighian bodies in the mesonephros of Peromyscus leucopus do not reach a sufficient stage of development to be considered as functional.

"The function of a renal corpuscle is to filter water and certain salts in solution from the blood." (Bremer, 1927). This function is accomplished by the inner layer of the capsule which in its mature form "consists of a single layer of flat epithelial cells which are intimately blended with each other and with the endothelium of the capillaries. The epithelial cells possess a clear cytoplasm and a flattened ovoid nucleus, which, being thicker than the body of the cell, produces a considerable bulging." (Jordan, 1927).

Bremer (1927) has described the development of this capsule briefly. "The younger glomeruli are covered by a tall columnar epithelium, the inner capsular layer, the height of the cells probably indicating preparation for rapid growth; the outer layer is of flat cells. The blood vessels push apart the cell bases of the inner layer and approach the surface, until only a thin plate-like layer of protoplasm covers the endothelium. The double nature of the thin layer overlying the blood stream of the glomerulus can occasionally be detected near the nuclei, where the plate and the endothelium separate. The cells between the capillaries become cuboidal as more and more of their substance is stretched over the endothelium. The capillary net is divided into lobules (usually five)

by deep clefts, plainly visible when the glomerulus is favorably oriented. The epithelial layer closely follows the lobulations, and thus presents a greatly increased surface."

In embryo No. 22, the outer wall of the capsule consists of greatly flattened cells with long ovoid nuclei which cause a bulging in the center of the cell. The cells of the inner wall are crowded very closely together, so that the nuclei protrude into the cavity between the two layers and have only a very thin layer of cytoplasm around them on the outer surface. The nuclei are slightly oval and the entire cell is stained very intensively by haematoxylin. These capsules are associated with tubules which are separated from the Wolffian duct and which are to a greater or lesser extent disintegrated at their proximal ends.

In the younger embryos, the cells of the outer layer are not so completely flattened while the inner layer is composed of more or less columnar cells with large oval nuclei. (Fig. 20).

Differentiation of the Tubule.

In those animals in which there is any indication of the mesonephros serving as an excretory organ, the development of the tubule is accompanied by a differentiation into a Bowman's capsule, a secreting and a collecting portion. Externally (from models of tubules) the differentiation is recognized by variations in size of the different parts of the tubule; internally the cellular structure is shown to be differentiated in a manner more or less similar to the comparable parts of the tubule of the metanephros.

Lewis describes the secreting portion of a tubule of the mesonephros from a human embryo of 16 mm. length. He says: "It is capacious, with wide lumen, and thick walls composed of somewhat elongated cells distinctly marked off from one another. These cells have conspicuous terminal bars, a frayed brush border, a superficial granular zone not deeply stained, and a still paler basal zone containing the oval nuclei. In a typical manner they show the histological features of secretory activity . . ." Concerning the next portion of the tubule, the collecting portion, he says: "the tubule becomes abruptly narrower, with a corresponding radical change in the histological

nature of its walls. The lumen is narrower, and the cells which are longer, with less cytoplasm stain deeply with haematoxylin.

In Peromyscus leucopus there is no differentiation of the tubule into secreting and collecting portions in so far as can be determined (Fig. 24). The diameter of the tubule is the same throughout its length, with the exception that in some tubules there is a slight constriction just before the tubule joins the Wolffian duct and in a part of those tubules which develop a Bowman's capsule, a similar constriction or neck is observable at the base of the capsule. The cells are similar in appearance throughout the length of the tubule and take a deep haematoxylin stain. They are more or less cuboidal, with very indistinct cell boundaries. The lumen of the tubule does not have a sharply defined margin as is the case with the lumen of the Wolffian duct. Nothing in the appearance of the cells of the tubules indicate any secretory activity.

Blood Supply of the Mesonephros.

In the human embryo branches from the aorta enter both the pronephros and the mesonephros and supply each tubule with a capillary. In the pig numerous arteries

from the aorta enter the mesonephros. In Peromyscus leucopus no capillaries even toward the mesonephros, are given off from the aorta before the degeneration of the organ is well under way. Even at this time the capillaries are few, three or four, and it is impossible actually to trace any of them into the mesonephros.

The blood supply of the mesonephros, then, differs from that of the human embryo, or of any other mammalian embryo in which it has been described, in that it appears to be entirely venous. The majority of papers, however, in describing the development of this organ^{in rodents} fail to mention the source of the blood supply, e. g., Weber, Bremer, Kerans, (loc. cit.).

The postcardinal vein makes its first appearance at the same time that the first mesonephric tubules begin to develop. When the urogenital fold is formed, the subcardinal vein develops in the ventral portion of the fold. The postcardinal communicates with the subcardinal by numerous branches and capillaries from the two veins ramify in the mesonephros.

Significance of Mesonephric Degeneration and
the Absence of an Allantoic Sack.

The early degeneration of the mesonephros in the human embryo and the absence of an allantoic sack have led the majority of authors to consider it non-functional as an excretory organ. Bremer (*loc. cit.*) thinks that possibly it is functional for a short period during the early embryonic development.

Because of the degenerate condition of the mesonephros and the absence of an allantois in the mouse, the rat, and the deer-mouse, there is no doubt that the mesonephros is non-functional in these forms also. We are led, therefore, to ask if there is any evolutionary connection between the conditions as found in these rodents and in man.

It does not seem that there is any possible relationship between the two cases. We find in man merely a case of very early degeneration of an organ which possesses all of the morphological qualifications necessary for functional activity, even though the essential glandular tissues, the tubules, are not so highly developed as in some other mammals. The absence of a lumen in the allantois, or at best the presence of a very small one, is the strongest argument against the possible functioning

of the mesonephros in the human embryo. Without the allantois there is no receptacle for the products secreted by the kidney.

The pattern of the tubules of the human mesonephros is more simple than that described for any other mammal since it is not even a complete reversed spiral. However, the tubule is differentiated into a secreting and a collecting portion and every tubule has a well developed Bowman's capsule and a glomerulus formed from an arterial capillary which is a branch from the aorta. ~~The Bowman's capsule and a glomerulus formed from an arterial capillary which is a branch from the aorta.~~ The Bowman's capsule is formed very early in the development of the tubule.

In contrast, we find the following conditions in the deer-mouse. There is a complete absence of an allantois. The tubule pattern is a very well developed double spiral but there is no differentiation of the tubule into secreting and collecting portions. The glomeruli are very rudimentary, probably incapable of functioning, and are found only on the tubules at the posterior end of the mesonephros. They are not formed until the tubule has reached its mature form. The blood supply to the capsules which are developed is entirely venous.

The character of the mesonephros in the teleost fishes and in the rodents appear to have several points in common. We find in each group species with aglomerular mesonephroi and others with mesonephroi which are entirely glomerular while between these two extremes are species which are to varying degrees only partly glomerular.

Within each group the renal corpuscles appear during the late stages of development of the renal tubules while in other mammals they are formed at an early stage in the development of the tubule.

Further it has been shown in each group that the excretory activity of the mesonephros may be taken over by another organ, in the case of the fish this organ is the gills; while in the mammals it is the placenta.

Edwards considers, however, that the aglomerular kidney in the fishes is more primitive than the glomerular. This view cannot be taken in connection with the mesonephroi of the rodents in which it is very apparent that the organ in this group is in a condition of retrogressive development.

In view of the fact that the conditions parallel each other so closely in the teleosts and the rodents it seems possible that we have here a case of parallel evolution of involution brought about possibly by similar causative factors which are still obscure.

The Metanephros.

The metanephros in its development appears to be similar in every way to that described for other mammals. In the last stage studied, embryo No. 22A, the tubules and glomeruli are developing but have not yet reached a stage of development which would enable the metanephros to function as an excretory organ.

The first generation of uriniferous tubules, using the terminology of Felix, is formed but is still in the developmental stages. Renal corpuscles are developing in connection with these tubules in the outer or cortical region. The cells of the outer wall of the capsule are flattened while those of the inner are still columnar and cuboidal in shape. The tubules are rather widely separated from each other by intertubular mesoderm.

The Gonads and the Müllerian Duct.

In Peromyscus leucopus the gonads are somewhat slower in their development than in Mus musculus, as nearly as can be determined from Brambell's (loc. cit.) description of the mouse. Sexual differentiation could not be determined in any except the last embryos of this series which have a length of eleven millimeters. In this stage the differentiation of the testis is more advanced than

that of the ovary. The testis cords are forming and the rete testis anlage appears as an unorganized mass of densely stained cells in the region between the testis and the mesonephros. Four tubules at the extreme anterior end of the mesonephros are still in communication with the Wolffian duct. However, no continuity between these tubules and the anlage of the rete testis has been established at this stage of development.

In the ovary, the primitive germ cells are scattered throughout the organ and have circles of epithelial cells around them indicating the beginning of the development of the follicles. None of the anterior tubules of the mesonephros are in communication with the Wolffian duct. The anlage of the rete ovarii has much the same appearance as that of the rete testis with the exception that the cells do not stain so deeply and the anlage as a whole is not so large. In general, differentiation and development in the ovary has not progressed as far as in the testis.

The development of the Müllerian duct agrees with other descriptions with the one exception that in the deer-mouse the ostium tubae abdominalis opens on the ventral side of the urogenital fold while in the embryo of man, and mammals generally, it opens on the dorsal

side. In embryos of 11.5 mm. length, these ducts have not quite reached the urogenital sinus.

The Urogenital Sinus and the Allantois.

In Peromyscus leucopus there is no indication, what-so-ever, of an allantoic diverticulum from the hind gut. Aside from this, the development of the urogenital sinus is comparable to that of other mammals. Although in Mus musculus an allantois is never developed sufficiently to extend into the umbilical cord, Weber (loc. cit.) does record a slight indication of such a structure and the formation of a urachis.

The absence of the allantois is of interest in connection with the development of the urinary bladder. In mammals, including man, it is generally considered to be of mixed origin, being derived from a portion of the cloaca, the part of the allantois inside of the body, and the portions of the Wolffian ducts which are absorbed into the wall of the bladder anlage as the ureters and mesonephric ducts are being separated from each other.

It is of interest here to note that there are in the different groups of vertebrates "bladder" organs which are in no way related to each other phylogenetically.

In the Amphibia the bladder is derived entirely from the ventral portion of the cloaca. According to Felix a bladder derived entirely from the allantois occurs temporarily in some birds while in the selanians the bladder is entirely mesodermal in its origin. The teleosts, ganoids, petromyzonts, snakes, crocodiles and mammals have bladders derived from the cloaca and the proximal portions of the primary excretory ducts.

Felix considers the human bladder to be derived from the ventral portion of the cloaca and the portion of the primary excretory ducts which are absorbed into its walls, but he does not think that any portion of the allantois stalk enters into its formation.

The urinary bladder in Peromyscus leucopus is derived from the dorsal portion of the urogenital sinus and the proximal portions of the Wolffian ducts. At the time when the urorectal septum is beginning to divide the cloaca into rectum and urogenital sinus, lateral evaginations form from the sides of the urogenital sinus immediately below the place where the Wolffian ducts end. These ducts do not enter the sinus at this time. Later the roof of the sinus between these evaginations is elevated, resulting in a long sack-like portion dorsal to the original urogenital sinus. It is

not until this sack, the anlage of the urinary bladder, is formed that the ends of the Wolffian ducts actually open into the cavity although they have been in contact with the wall throughout almost the entire period of development of the mesonephros.

The mesonephros, then, cannot have been functional as an excretory organ because there would have been no means of escape for the excretory products, since by the time that the mesonephric ducts open into the anlage of the urinary bladder, the entire glandular portion of the organ has disintegrated.

In its further development, in so far as it has been observed in this series of embryos, the urorectal septum continues to separate the rectum from the urogenital sinus until only a small duct, the urogenital duct, remains, to connect the two portions. The original portion of the urogenital sinus ceases to develop except as it continues to grow with the general growth of the body. It is connected directly along its ventral side with the external body ectoderm throughout its length, thus forming the urogenital membrane. The anlage of the urinary bladder continues to develop, pushing forward into the mesoderm attached to the ventral body wall at its anterior end. It absorbs the basal portion of the Wolffian duct

which thus becomes separated from the ureter. The development of the derivatives of the urogenital sinus was not followed further in this study.

CONCLUSIONS.

1. The mesonephros in the embryos of Peromyscus leucopus attains a degree of development slightly greater than is reached in the mouse and the rat, but not so great as in the guinea pig.
2. It never becomes functional as an embryonic excretory organ.
3. The essential tissues of the mesonephros, the tubules, are never differentiated into secreting and collecting portions, although the tubules acquire a pattern, the reversed spiral, comparable to that of many other mammals in which the mesonephros is presumably functional. There is no differentiation within the tubule of the various types of cells found in a functioning mesonephros.
4. Bowman's capsules do not develop on the tubules until the last stage of development of the mesonephros and then, they develop only on those at the posterior end of the organ. When present, they are very rudimentary and comparable to the earliest stages of development of capsules in other mammals. Three or four more-or-less mature capsules are to be found only during the final degenerative stages of the mesonephros and after

all of the tubules have become separated from the Wolffian duct.

5. Capsules were found to be invaginating on a few tubules without a blood capillary in contact with them.
6. There is apparently no arterial blood supply to the organ during its developmental period. The posterior cardinal vein appears at about the time that the first mesonephric tubules begin to develop and this vein and the sub-cardinal supply the organ with blood.
7. Rudimentary glomeruli are apparently developed from venous capillaries.
8. The Wolffian duct reaches the cloaca at a very early stage of development of the mesonephros but does not open into the urogenital sinus until the mesonephros has almost reached its final stage of degeneration.
9. No trace of an allantois is ever evaginated from the hind gut.
10. The urinary bladder is derived from the dorsal and lateral portions of the urogenital sinus and the proximal portions of the primary excretory ducts.

11. There is no evolutionary connection between the degenerate condition of the mesonephros as found in the human embryos and that of the embryos of the deer-mice.
12. There may be some causative relationship between the loss of the glomeruli in certain of the teleost fishes and in some of the rodents including Peromyscus leucopus.

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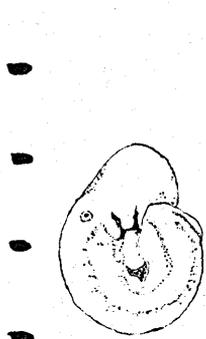
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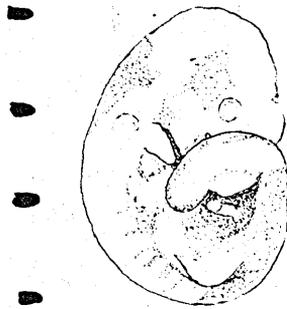
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Plate I.

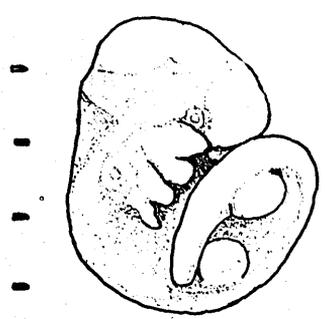
- Fig. 1. Embryo No.40. G.L. 2.1 mm.
Fig. 2. Embryo No.434. G.L. 3.1 mm.
Fig. 3. Embryo No.34. G.L. 4.2 mm.
Fig. 4. Embryo No.27. G.L. 5.2 mm.
Fig. 5. Embryo No.435. G.L. 6.1 mm.



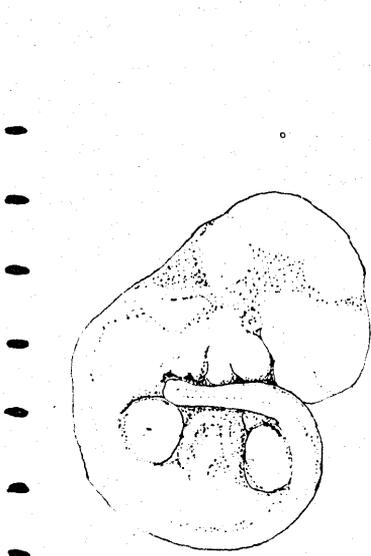
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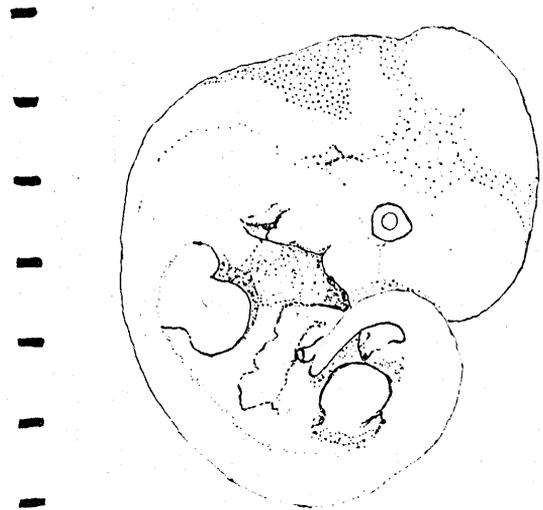
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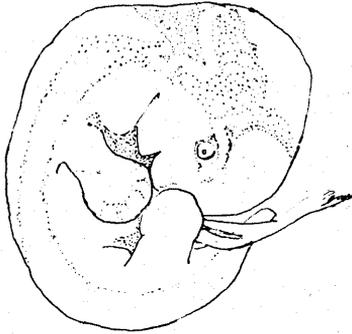
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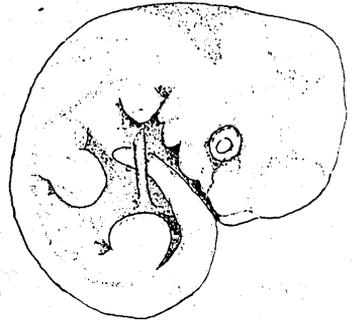
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Plate II.

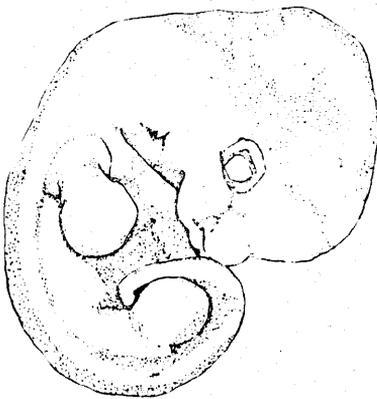
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| Fig. 6. | Embryo No.42. | G.L. 6.4 mm. |
| Fig. 7. | Embryo No.230. | G.L. 6.5 mm. |
| Fig. 8. | Embryo No.21. | G.L. 8 mm. |
| Fig. 9. | Embryo No.22. | G.L. 11.5 mm. |



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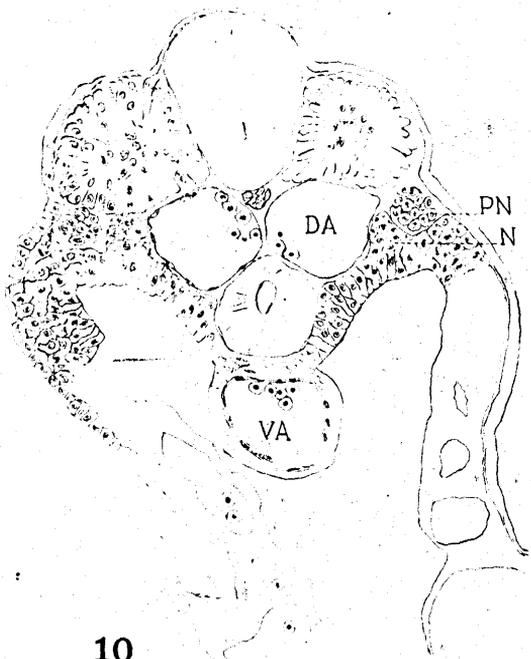
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Plate III.

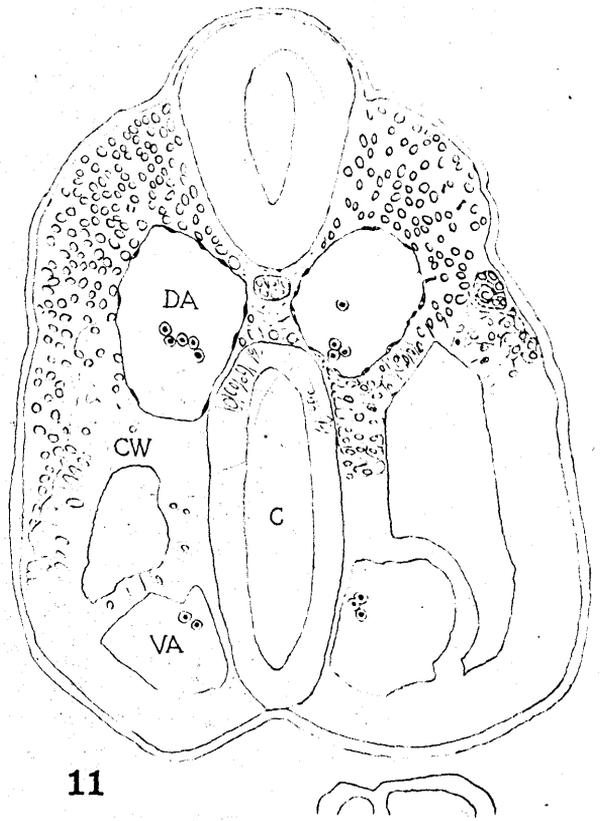
- Fig. 10. Cross section through the trunk region of embryo No.40.
- Fig. 11. Cross section through the same embryo thirty sections posterior to fig.10.
- Fig. 12. Embryo No.34. To show the intimate relationship between the end of the Wolffian duct and the wall of the cloaca. The duct ends a blind tube three sections posterior to the one from which this drawing was made. Drawn x 1000 dias.
- Fig. 13. Cross section through embryo No.34. This is the same section from which Fig. 12 was made.

Abbreviations.

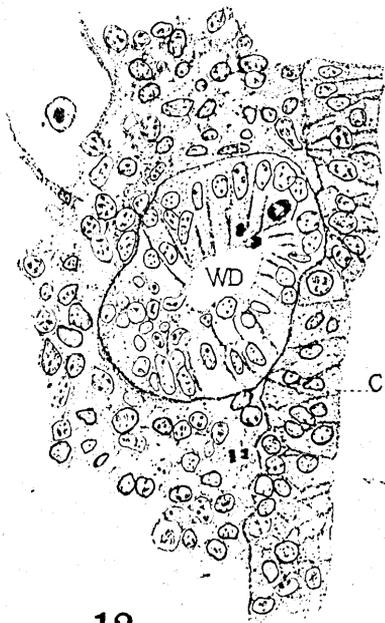
C	Cloaca.
CW	Wall of coelom.
DA	Dorsal Aorta.
VA	Ventral artery.
N	Nephrotome.
PN	Pronephric duct.
WD	Wolffian duct.



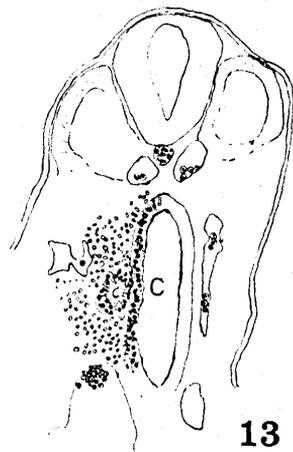
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Plate IV.

- Fig. 14. Cross section through the anterior trunk region of embryo No. 434. This is the earliest stage in which the post cardinal vein appears.
- Fig. 15. Cross section through the anterior trunk region of embryo No. 34. The post cardinal vein connects with the sub cardinal on the left side of the illustration but does not on the right side.
- Fig. 16. Embryo No. 280. Cross section through the fifteenth tubule of the mesonephros. The developing gonad is shaded slightly darker than the mesonephros.
- Fig. 17. Embryo No. 21. Cross section through the sixteenth tubule of the mesonephros.

Abbreviations.

DA	Dorsal aorta.
PC	Post cardinal vein.
SA	Segmental artery.
MA	Mesenteric artery.
WD	Wolffian duct.

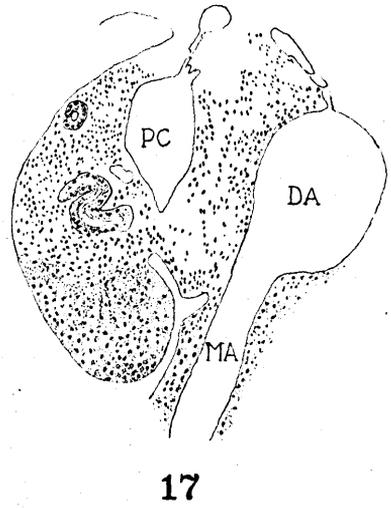
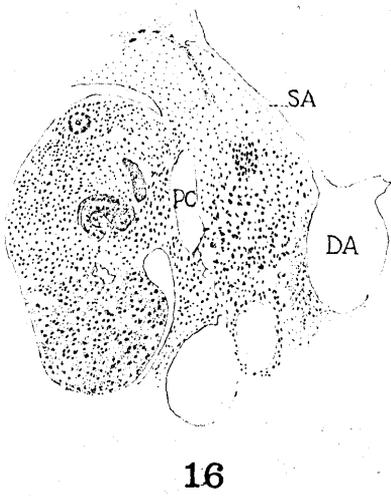
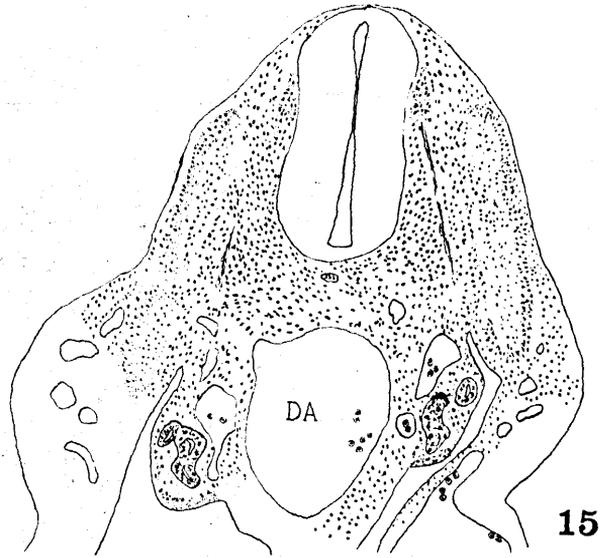
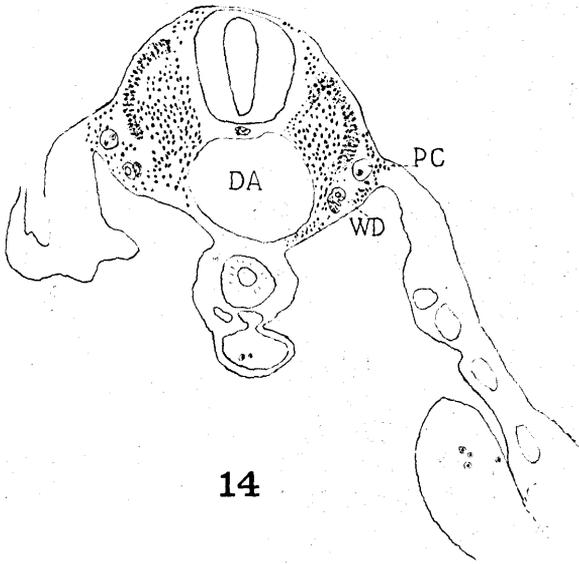
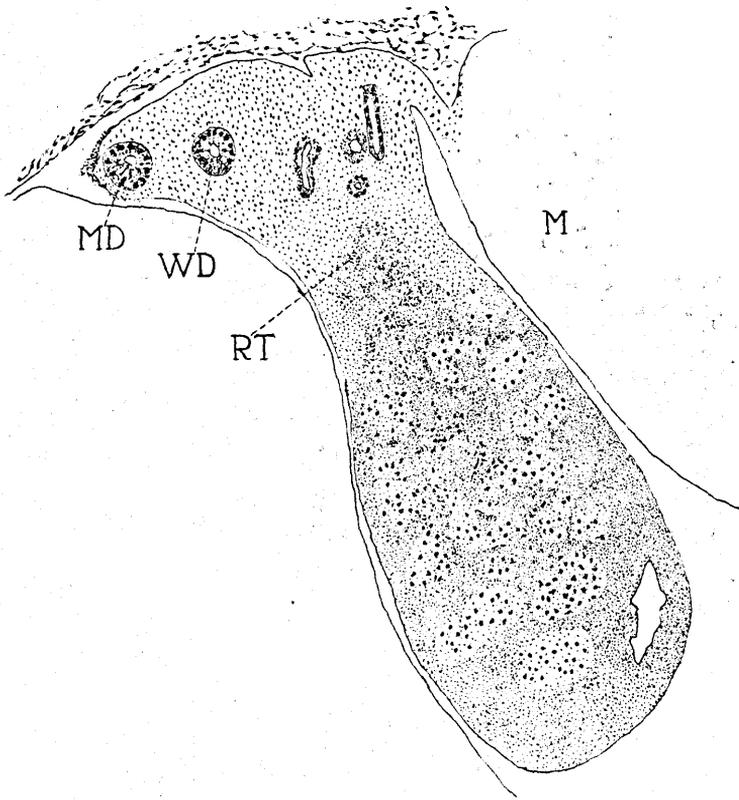


Plate V.

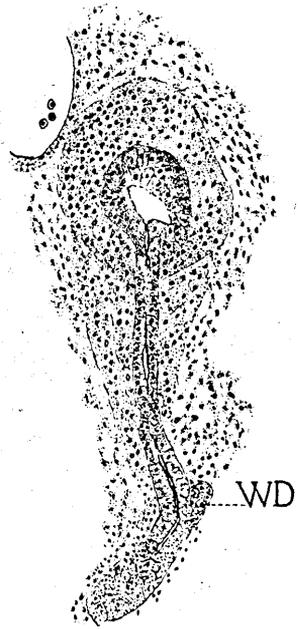
- Fig. 18. Embryo No.22A. Cross section through the mesonephros and testis.
- Fig. 19. Embryo No.435. Section through the length of the ureter and cross section of the vesicular portion.
- Fig. 20. Embryo No.280. Section through a glomerulus. The capillary shown is from the post cardinal vein.
- Fig. 21. Embryo No.22A. Section through the urogenital fold showing the funnel-shaped pit which forms the ostium of the Müllerian duct.

Abbreviations.

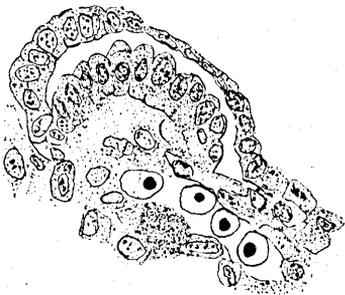
L	Liver.
M	Metanephros.
MD	Müllerian duct.
WD	Wolffian duct.
RT	Rete testis anlage.



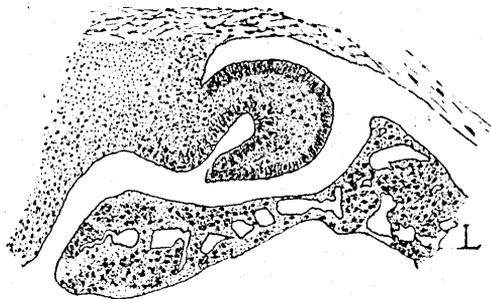
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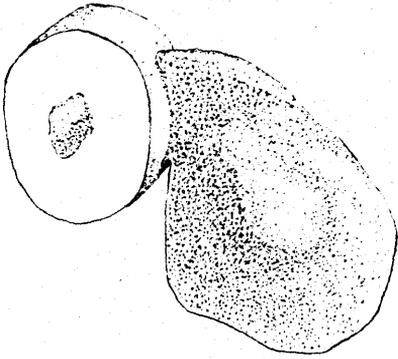
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Plate VI.

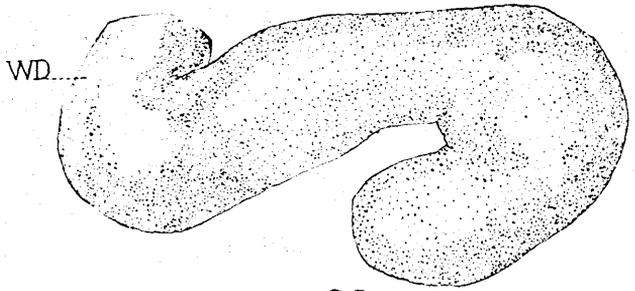
Fig. 22. Embryo No.34. Drawing of a wax reconstruction of the eleventh mesonephric tubule.

Fig. 23. Embryo No.27. Drawing of a reconstruction of the first mesonephric tubule.

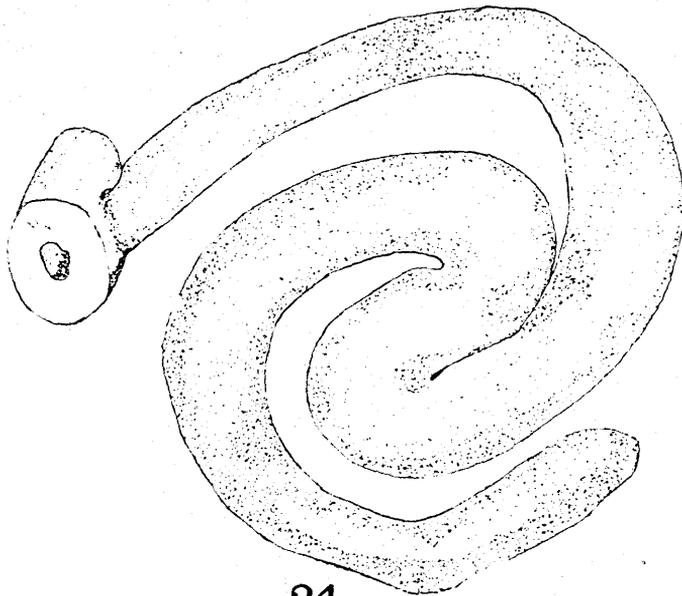
Fig. 24. Embryo No.280. Drawing of a reconstruction of the ninth mesonephric tubule.



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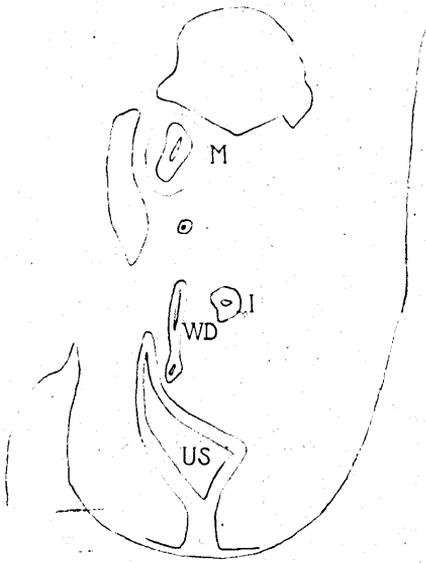
Plate VII.

Camera lucida drawings to show the relationship of the various ducts etc. to the urogenital sinus.

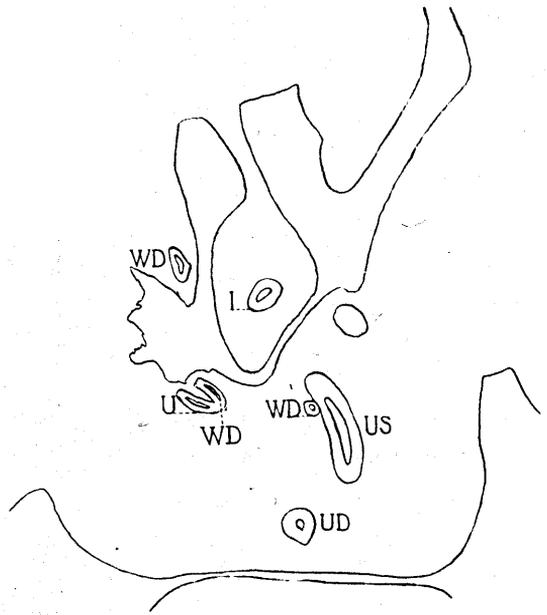
- Fig. 25. Embryo No.42. End of Wolffian duct in contact with the evagination from the side of the urogenital sinus.
- Fig. 26. Embryo No.280. End of Wolffian duct in contact with the evagination from the side of the urogenital sinus.
- Fig. 27. Embryo No.21. Wolffian duct opening into the posterior end of the evagination from the urogenital sinus.
- Fig. 28. Embryo No.22A. The end of the Müllerian duct at (x). The Müllerian duct crosses over to the inner side of the Wolffian duct in this region.
- Fig. 29. Embryo No.22A. The Wolffian duct opening into the urogenital sinus.
- Fig. 30. Embryo No.22A. The ureter opening into the urogenital sinus.

Abbreviations.

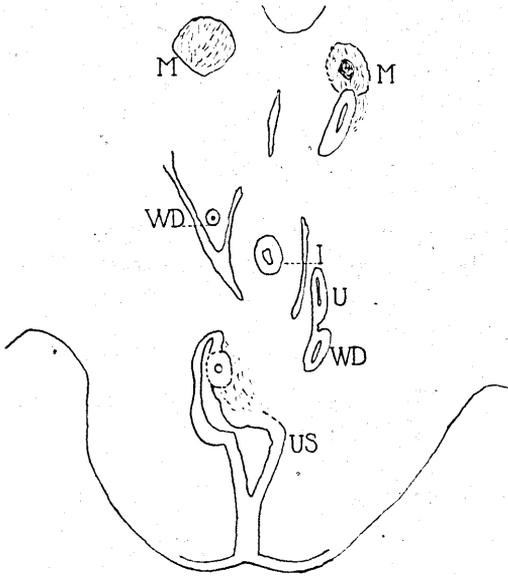
I	Intestine.	U	Ureter.
M	Metanephros.	UD	Urogenital duct.
MD	Müllerian duct.	US	Urogenital sinus.
P	Pancreas.	WD	Wolffian duct.



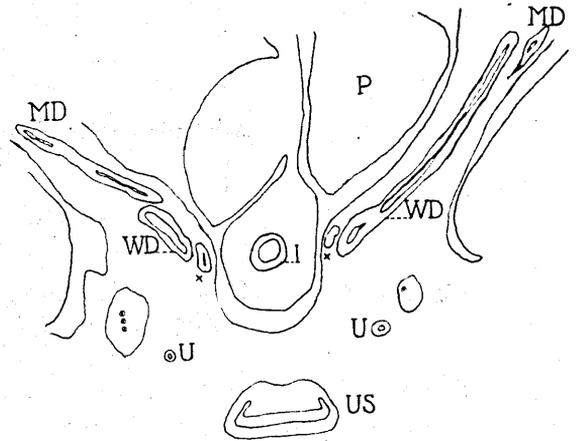
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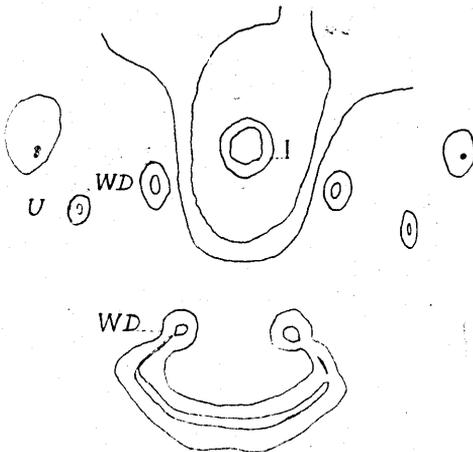
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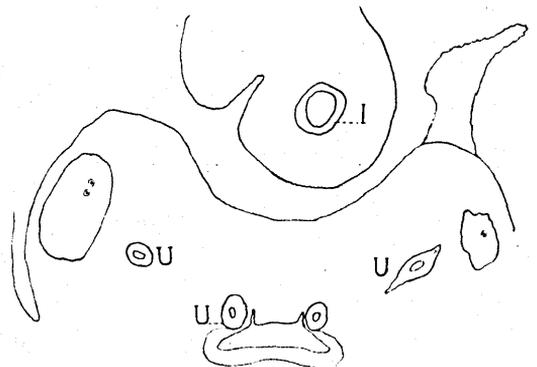
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