THE POSTNATAL DEVELOPMENT OF THE KIDNEY IN THE ALBINO MOUSE

by

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STATEMENT BY AUTHOR

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ABSTRACT

Tracing the postnatal development of the albino mouse kidney from the newborn to the 22 day old reveals no histologically discernible maturation after 17 days. Many developing renal corpuscles can be seen in the outer cortex during the first week of life. At seven days of age the proximal tubules have brush borders and basal striations. The epithelium of the papillary ducts changes from tall columnar in the newborn to high cuboidal in the 17 day old kidney, and the papilla increases in length for 17 days. At birth large areas of undifferentiated tissue are present in the outer medulla and persist in decreasing amounts through the sixteenth day after birth. The undifferentiated mesenchyme changes first into bipolar cells, then epithelioid cells, and finally into primitive tubules. At 17 days all undifferentiated tissue has disappeared and the tubules can be clearly identified. The ratio of the length of medulla to length of kidney more than doubles between birth and 17 days.

INTRODUCTION

The development of the metanephric kidney may continue after birth for varying amounts of time depending on the species. The nephrons and the system of collecting tubules and ducts arise independently. The renal pelvis, the calyces, the collecting ducts, and finally the collecting tubules come from repeated branching of the metanephric bud, a diverticulum of the old mesonephric duct. The nephrons develop through differentiation of a compact mesenchyme, the metanephric blastema, which forms a cap over the branching ends of the metanephric bud. The nephrons and the arched collecting tubules eventually connect to complete the functional units of the kidney.

Most of the histological studies of postnatal development have been done on the rat kidney. During the first few weeks after birth new nephrons continue to form in the rat kidney cortex (Kittelson, 1917; Arataki, 1926). The mature rat kidney contains almost three times as many glomeruli as the newborn, and the diameter of the glomerulus approximately doubles between birth and maturity (Arataki, 1926). Renal function also is incompletely developed at birth. The newborn rat excretes a dilute urine of relatively unvarying composition and volume (Falk, 1955).

The mouse, like several other mammals, including the rat and man, is born with an incompletely differentiated kidney. At birth the mouse kidney is a pale, almost colorless translucent body, but becomes progressively darker red and opaque throughout the first two weeks of life. Some glomeruli are fully formed before birth and the macula densa and juxtaglomerular cells appear in fetal mice of 16 and 17 days gestation (Kaylor and Carter, 1967).

Two well-known characteristics of the proximal tubules of the kidney cortex as seen with the light microscope are the brush border and basal striations. The electron microscope has shown the brush border to be composed of long, tightly packed microvilli at the apical ends of the cells, and the basal striations to be composed of mitochondria lined up between long infoldings of the cell membrane perpendicular to the basement membrane (Sjostrand and Rhodin, 1953; Pease, 1955). In an electron microscope study of the deeper layers of mouse kidney cortex, Clark (1957) found that both of these characteristics appear during the first two weeks after birth.

The electron microscope study was confined to small areas of the inner cortex and ignored the peripheral cortex and medulla, but Clark (*ibid.*) stated that the two week old mouse kidney is histologically indistinguishable from that of the adult mouse. While making routine histological preparations of mouse kidneys, I found this observation to be untrue.

The purpose of the present light microscope study is to describe the postnatal changes in the mouse kidney and to determine at what age the kidney becomes histologically indistinguishable from that of the adult.

MATERIALS AND METHODS

Litters of albino mice were sacrificed with ether or chloroform at the following postnatal stages: newborn, seven days old, and daily from 14 through 23 days of age. A day old mouse and a breeding age female were also sacrificed for comparison. The kidneys were placed in Bouin's fixative, then transfered to 70% ethanol to which several drops of saturated aqueous lithium carbonate had been added to remove the picric acid. The kidneys were dehydrated in 95% and 100% ethanol, cleared in toluene, and embedded in paraplast. Serial sections were cut at 10 microns and stained with Mayer's acid haemalum (Lillie, 1954) and eosin.

RESULTS

Median sagittal sections of the newborn mouse kidney average 2.45 mm in length. The kidney is covered by a very thin layer of attenuated mesothelial cells which is rarely more than one cell layer thick. The capsule is very closely applied to the cortex, and there are no fibers visible in it at this time.

The outer cortex is extremely basophilic, containing many developing renal corpuscles (Fig. 1 and 2). These corpuscles can be seen in all stages of development, from the expanding end of the renal tubule just beginning to form Bowman's capsule, to advanced stages containing the glomerulus. Figure 3 shows a renal corpuscle with a well developed macula densa. What appears to be developing juxtaglomerular cells occur between the basement membrane of the macula densa and the basement membrane of the parietal layer of Bowman's capsule. Mammalian glomeruli have been shown to have a long and a short axis, appearing as ellipsoids in histological sections (Munkacsi and Palkovits, 1965). Measured along the long axis, fully formed glomeruli in the new born mouse kidney average 72.5 microns in diameter.

Figure 1. Newborn mouse kidney. c, cortex with developing renal corpuscles; ms, stellate mesenchyme; p, papilla with ducts of Bellini. Magnification 26.8X.

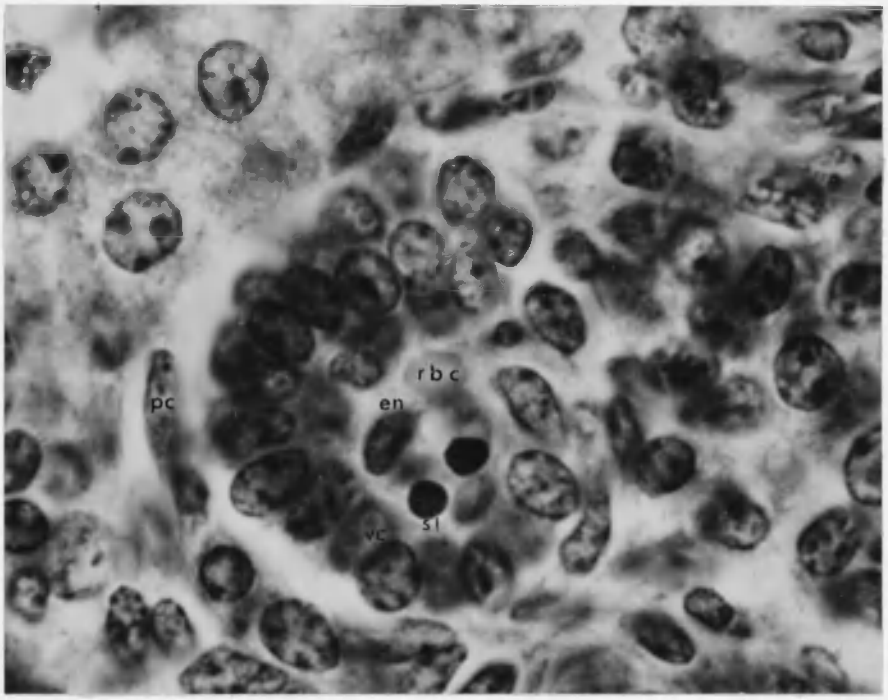

Figure 2. Developing renal corpuscle in the peripheral cortex of the newborn mouse kidney. pc, parietal Bowman's capsule; vc, visceral Bowman's capsule; en, endothelial cell of glomerulus; sl, small lymphocyte; rbc, red blood cell. Magnification 2550X.

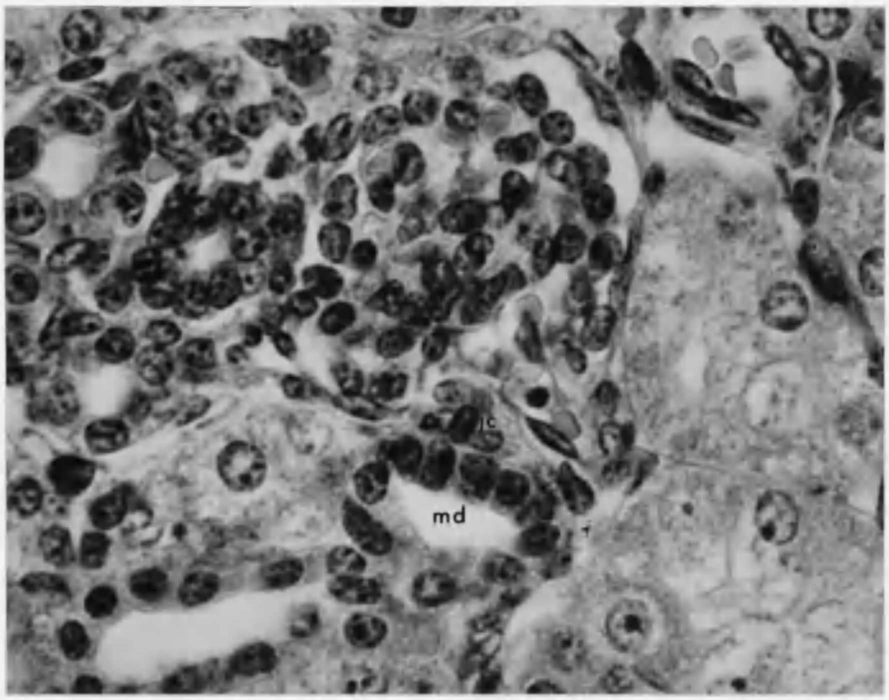

Figure 3. Fully formed renal corpuscle from newborn mouse kidney. md, macula densa; jc, juxtaglomerular cells. Magnification 1180X.

The inner cortex contains eosinophilic proximal convoluted tubules with a bright pink refractory border. This border does not have the faintly striated appearance of the fully differentiated brush border. No basal striations are present.

The cortex is penetrated by several medullary rays radiating from the medulla and containing straight collecting tubules. A few thin segments of the loop of Henle are present. The loop of Henle is the last part of the nephron to form (Huber, 1905).

Many mitotic figures are present in the basophilic outer cortex, but they also occur in the inner cortex in the many slightly basophilic and relatively undifferentiated tubules. A few mitotic figures occur in some tubules which have already developed some eosinophilia.

The outer medulla is composed of large areas of stellate mesenchyme, with a few scattered basophilic tubules (Fig. 4).

The papilla is very short (Fig. 1). The length of the medulla, measured from the junction with the cortex to the tip of the papilla, averages 0.7 mm. A few ducts of Bellini, or papillary ducts, are present. The papillary ducts are composed of a tall columnar epithelium showing some eosinophilia. The eosinophilia is particularly evident along the free surface where the cytoplasm stains a much

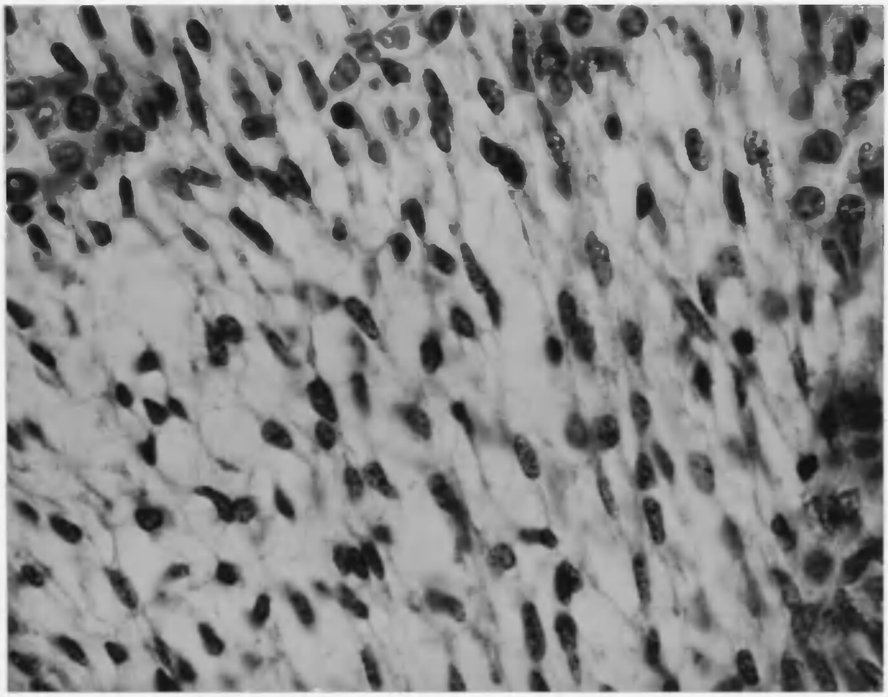

Figure 4. Stellate mesenchyme from the outer medulla of newborn mouse kidney. Magnification 1160X.

deeper pink. The ducts of the inner medulla are composed of a more basophilic cuboidal epithelium, and contain some mitotic figures. The tip of the papilla is covered with tall columnar epithelium which becomes cuboidal farther away from the tip, and then squamous near the renal pelvis. The squamous epithelium lines most of the rest of the renal pelvis but becomes transitional near its junction with the ureter. A few mitotic figures occur in the squamous or cuboidal epithelium of the renal pelvis.

The day old mouse kidney is very similar to that of the newborn. The capsule is a single layer of mesothelial cells. The outer cortex is very basophilic and contains many developing nephrons and many mitotic figures. Large areas of stellate mesenchyme remain in the inner cortex and outer medulla. A number of eosinophilic proximal and distal tubules are present and many small round tubules consisting of four or five slightly basophilic cells occur in the inner cortex. Mitotic figures are rather common in these tubules. The papilla of the day old mouse is slightly larger than that of the newborn, and the ducts of Bellini can now be traced well into the medulla. The cell boundaries of the columnar cells are visible, but they are not as distinct as they are in the adult.

A large duct approximately 300 microns in length and believed to be a remnant of the mesonephric duct appears near the hilus (Fig. 5). This duct descends from the cortex, but is not continuous with any of the kidney tubules. The wall is composed of simple columnar epithelium which has a prominent basement membrane. The columnar cells are large, pale and hypertrophic in appearance with small round nuclei near the lumen. The duct passes close to the ureter and develops an S- shaped curve before ending blindly. The sigmoid curve appears to be a distortion caused by pressure from the growing ureter.

Median sagittal sections of the seven day old kidney average 4.25 mm. The capsule has a thin layer of collagenous fibers and is about seven to 10 microns thick.

Renal corpuscles continue to develop in the outer cortex, but the peripheral basophilic area is narrower and stains purple rather than dark blue as it does in the newborn (Fig. 6). Mitotic figures are numerous in this area. Although the renal corpuscles of the seven day old mouse may have a fairly large glomerulus, some of these corpuscles still have a low cuboidal parietal epithelium rather than squamous as in the fully formed corpuscles. The average diameter of the fully formed glomerulus is 81.3 microns.

At seven days the cortex contains many more eosinophilic proximal tubules, and they extend farther toward

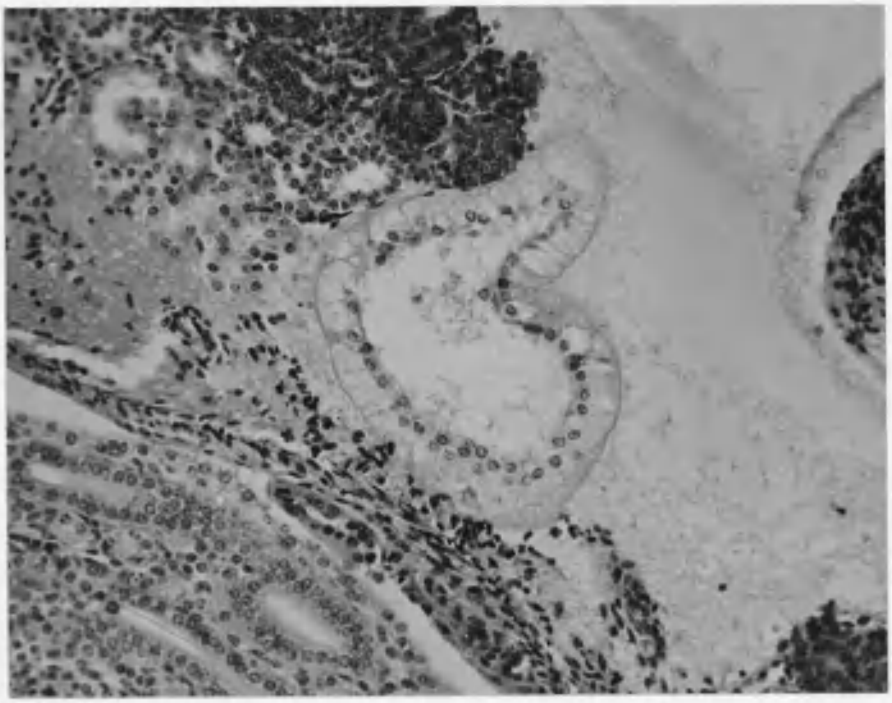

Figure 5. Remnant of the mesonephric duct near the hilus of a day old mouse kidney. Magnification 268X.

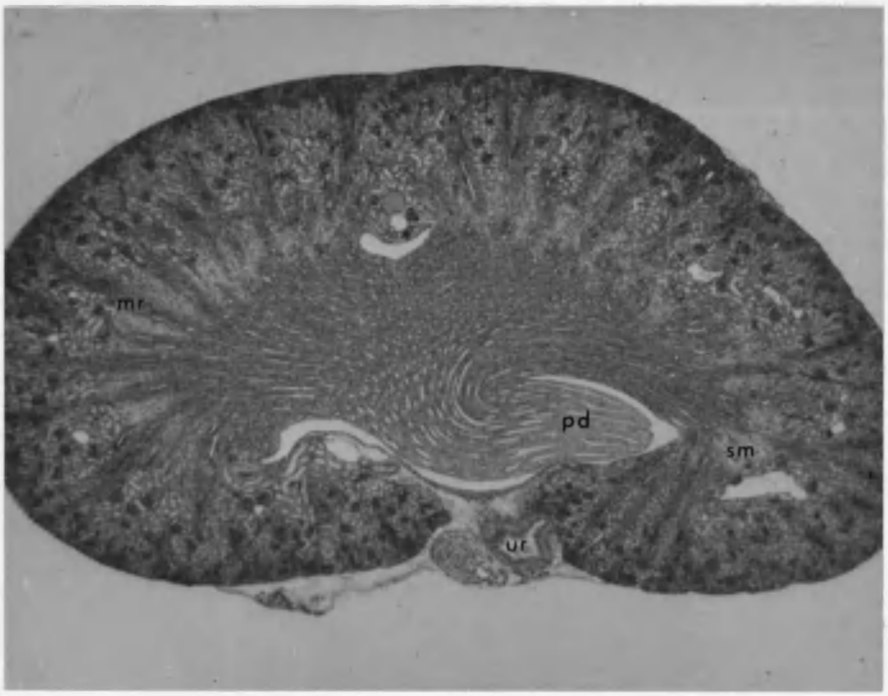

Figure 6. Seven day old mouse kidney. mr, medullary ray; sm, stellate mesenchyme; pd, papilla with papillary ducts; ur, ureter. Magnification 26.8X.

the periphery than in the newborn and day old mouse kidneys. Basal striations and brush borders are evident in these tubules.

The outer medulla contains large areas of mesenchyme and many capillaries (Fig. 7), but there are more medullary rays present than in the day old kidney. The mesenchymal cells between the bases of the medullary rays are elongating and becoming bipolar with an orientation parallel to each other and perpendicular to the medullary rays. Some stellate cells are still present at seven days, however. Small pale bluish tubules are scattered throughout the mesenchyme of the outer medulla.

The medulla averages 1.93 mm in length in the seven day old mouse, and the papilla is much longer than in the day old kidney (Fig. 6). The papillary ducts extend well into the medulla. The cells of the ducts at the tip of the papilla are somewhat columnar, but in the inner medulla they are cuboidal with fairly distinct cell boundaries and their free surfaces bulge into the lumen as in the adult. The cells covering the tip of the papilla are columnar, but shorter and more eosinophilic than in the day old kidney. Mitotic figures are not common in the papilla but are fairly numerous in the medulla.

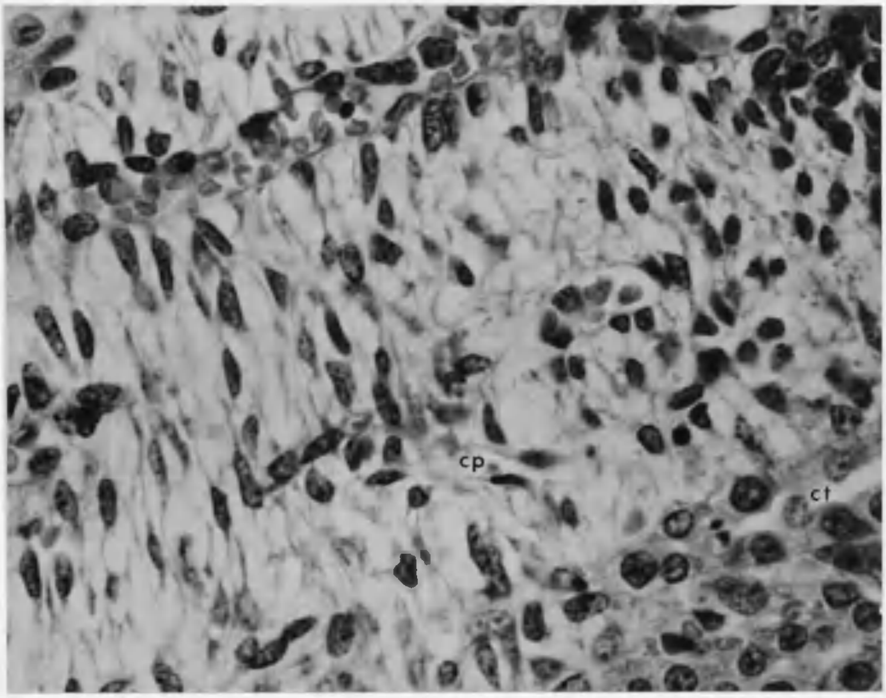

Figure 7. Mesenchyme from outer medulla of seven day old mouse kidney. cp, capillary; ct, collecting tubule in medullary ray. Magnification 1180X.

There is now a layer of smooth muscle underlying the transitional epithelium of the renal pelvis, and the muscle layer surrounding the ureter has become very thick.

At 14 days the length of the median sagittal section has increased to 5.63 mm. The capsule is much more fibrous than at seven days and has fibroblasts four or five cells deep in the thicker areas. The layers of fibers and fibroblasts may be more than 20 microns thick.

The cortex is now filled to the outer edge with eosinophilic tubules and contains many renal corpuscles (Fig. 8). Mitotic figures are fairly numerous in the tubules, and a few corpuscles with a cuboidal parietal layer are still present. The diameter of the glomerulus has increased to 86.7 microns.

There is a large pale blue-staining area in the outer medulla containing cross sections of many small primitive tubules composed of only three or four cells with large round nuclei. Many of the tubules have no lumen. The cells between the tubules are so epithelioid that it is difficult to distinguish tubules from interstitial cells (Fig. 9). Many mitotic figures occur in the epithelioid cells and primitive tubules. In a few spots the outer medulla contains masses of epithelioid cells which do not appear to be arranged in tubules.

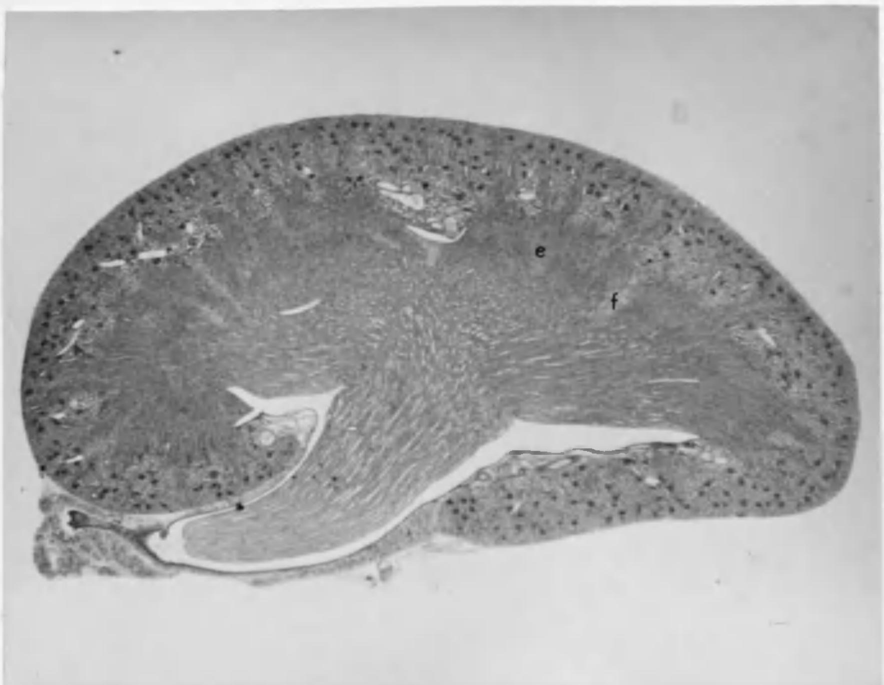

Figure 8. Fourteen day mouse kidney. f, small patches of fibroblasts;, e, blue-staining area containg epithelioid cells. Magnification 26.3X.

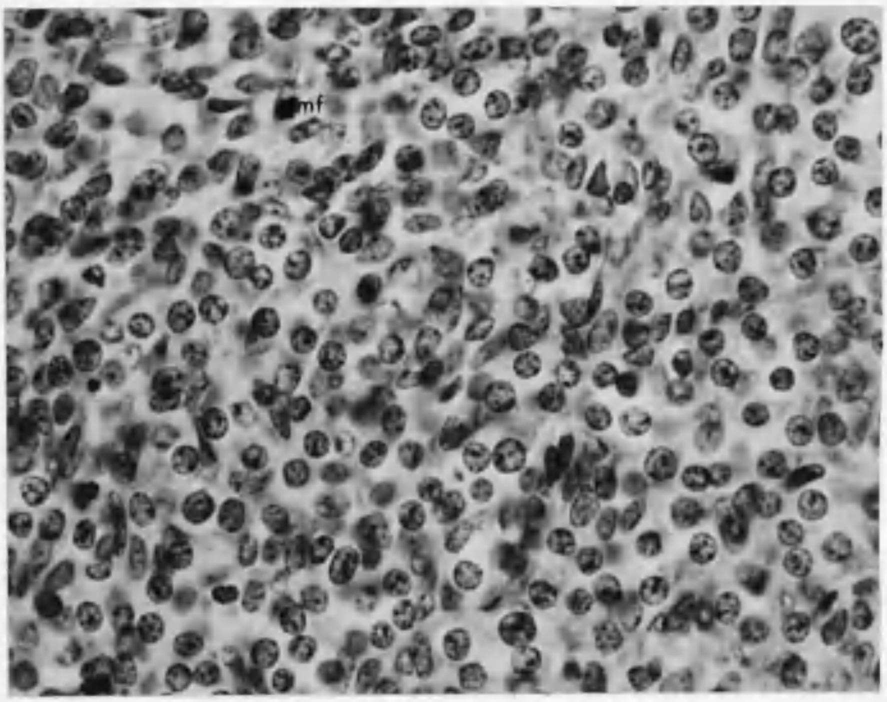

Figure 9. From the medulla of 14 day old kidney. Epithelioid cells not arranged in tubules yet. mf, mitotic figure. Magnification 1180X.

A few small areas containing fibroblasts and capillaries remain in the outer medulla (Fig. 8), and most of the fibroblasts are oriented parallel to each other and perpendicular to the medullary rays (Fig. 10).

As can be seen in Figure 8, the renal papilla of the 14 day old mouse is much longer than that of the seven day old mouse. The medulla averages 3.38 mm. The tip of the papilla is covered with a low columnar or high cuboidal eosinophilic epithelium. The ducts of Bellini near the tip of the papilla have a slightly taller epithelium than those of the adult, but the free surfaces of the cells are quite convex and cell boundaries are more distinct than in earlier stages. The surface of the rest of the papilla is covered by a low cuboidal or high squamous epithelium with regularly spaced nuclei. The renal pelvis is lined with a low squamous epithelium with elongated and irregularly spaced nuclei. The epithelium changes from squamous to transitional as it approaches the ureter, and the underlying layer of smooth muscle becomes thicker.

The capsules of the 15 and 16 day old kidneys contain slightly heavier fibers, but are not much more than 20 microns thick. The capsule does not seem to gain much in total thickness up to 22 days, at which time it is still not as thick and dense as that of the adult. The adult capsule is quite dense, and may be as much as 30 microns thick.

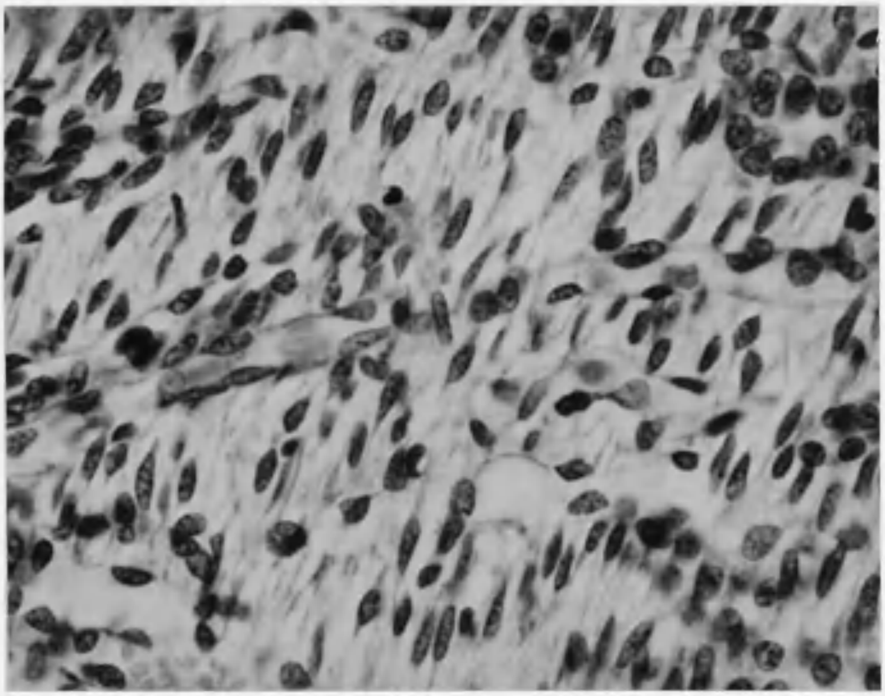

Figure 10. Small area in outer medulla of 14 day old kidney containing fibroblasts and capillaries. Note parallel orientation of most of the fibroblasts. Magnification 1180X.

At 15 days there are many mitotic figures in the cortex. Large blue-staining areas in the outer medulla contain closely packed epithelioid cells and a few primitive tubules (Fig. 11). Between the bases of the medullary rays there are a few small areas that are less densely cellular and contain fibroblasts and a few small areas that are less densly cellular and contain fibroblasts and a few capillaries (Fig. 12). Thin segments of the loop of Henle are more evident in the medulla. No mitotic figures were found in the papilla of the 15 day old, nor in any of the later stages.

At 16 days the pale blue-staining areas of the medulla are filled with tightly packed primitive tubules which still contain mitotic figures (Fig. 13). The largest patch of fibroblasts found in any of the 16 day kidneys is seen in Figure 14. At 16 days the epithelium of the ducts of Bellini is tall cuboidal as in the adult, but the cell boundaries are not as distinct.

At 17 days the length of the kidney has increased to 5.68 mm, the length of the medulla to 3.65 mm, and the average diameter of the glomerulus has increased to 89.4 microns (Fig. 15). All undifferentiated tissue has disappeared (Fig. 16). The tubules can be clearly identified and the cell boundaries in the ducts of Bellini are quite distinct.

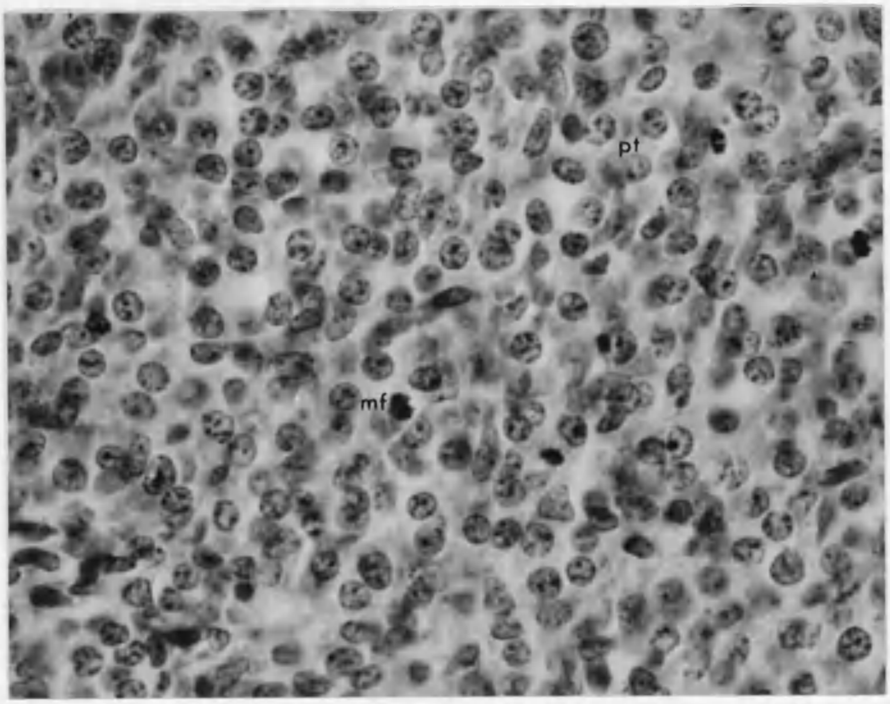

Figure 11. Epithelioid cells from the medulla of a 15 day kidney. pt, primitive tubule; mf, mitotic figure. Magnification 1180X.

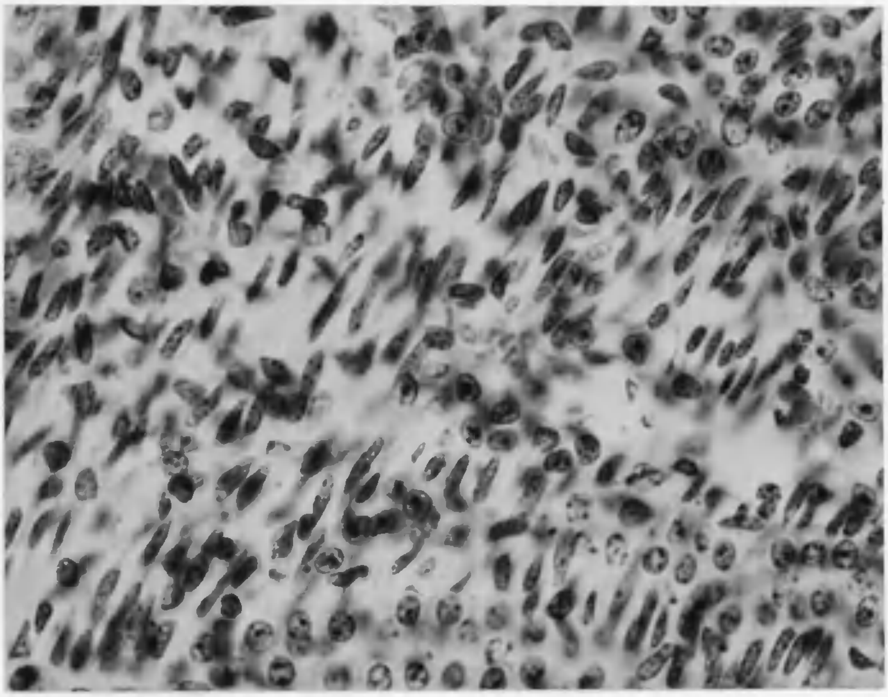

Figure 12. Fifteen day kidney. Small patch of fibroblasts in the medulla. Magnification 1180X.

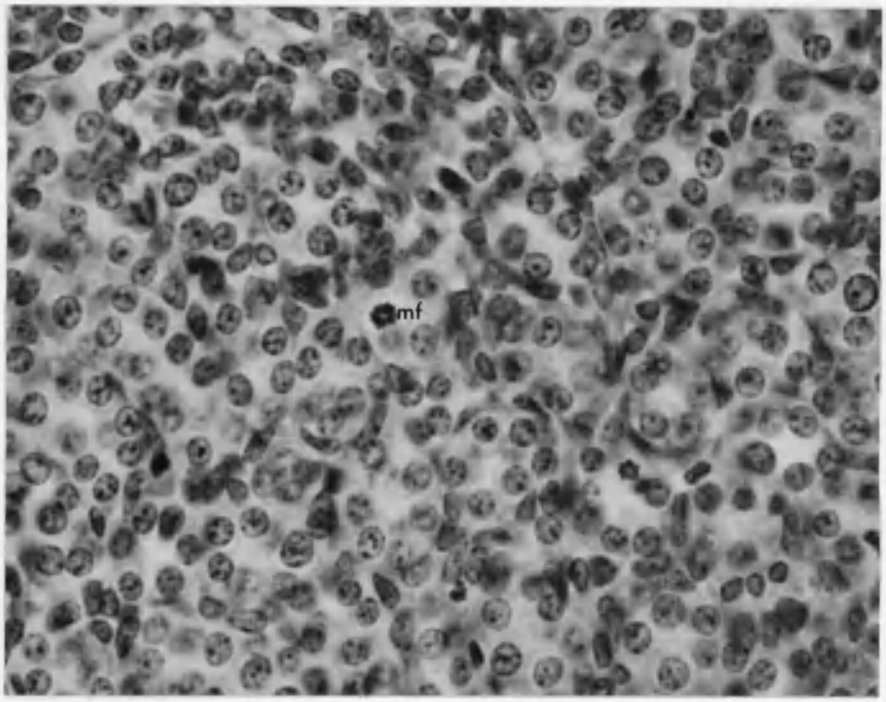

Figure 13. Primitive tubules from the medulla of a 16 day kidney. mf, mitotic figure in a tubule. Magnification 1180X.

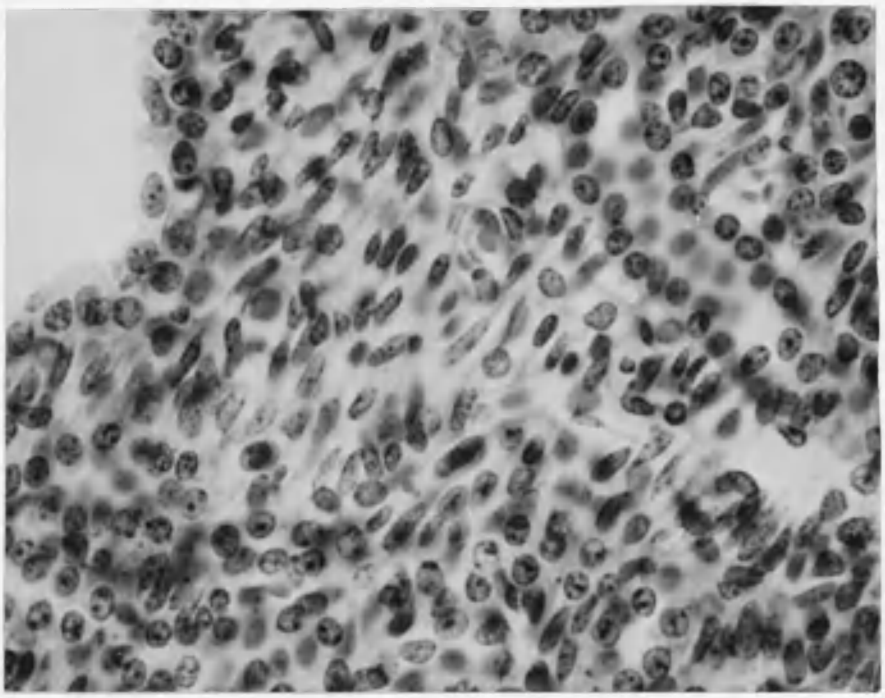

Figure 14. Sixteen day kidney. Tiny patch of fibroblasts in the outer medulla. Magnification 1180X.

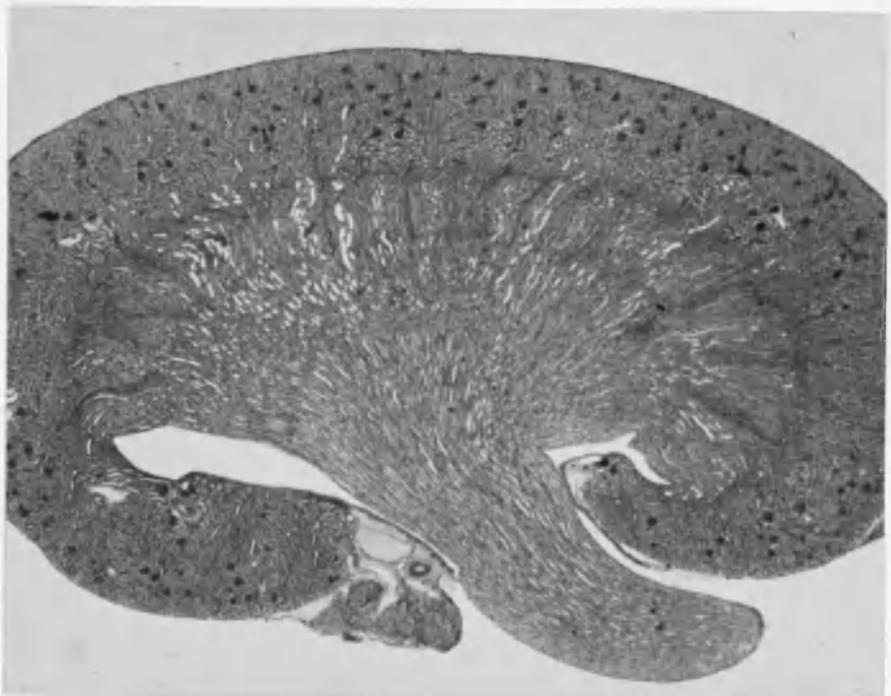

Figure 15. Seventeen day old mouse kidney. Magnification 26.8X.

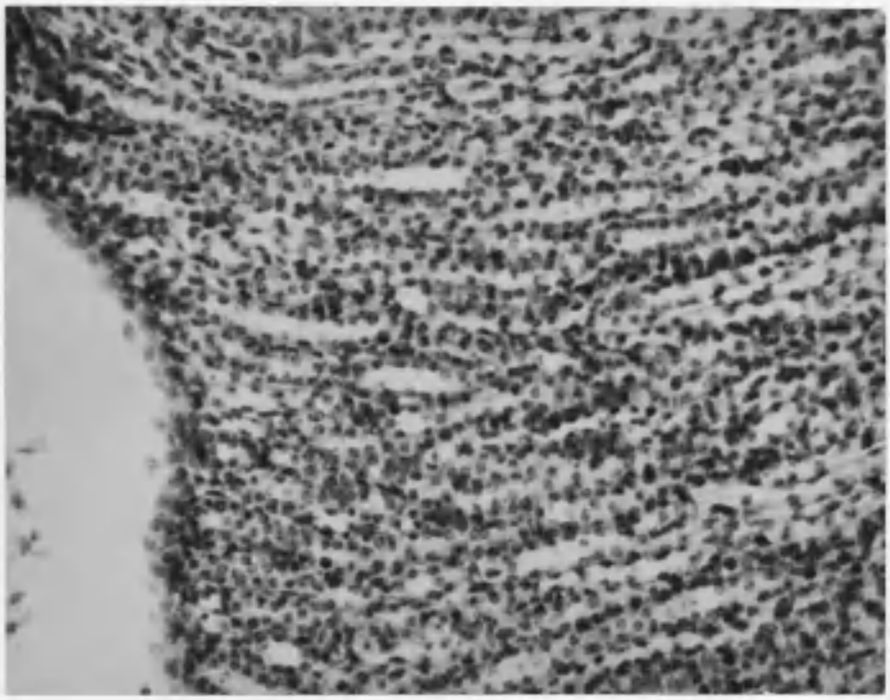

Figure 16. Outer medulla of the 17 day kidney. Includes the area corresponding to that shown in Fig. 14. Magnification 263X.

DISCUSSION

With the disappearance of the undifferentiated tissue at 17 days the mouse kidney has become histologically mature. The characteristics of the tubules and ducts are identical with those of the adult. Though the glomerulus of the 17 day old mouse kidney is slightly smaller than that of the adult (89.4 microns at 17 days and 91.7 microns in the adult) it is otherwise indistinguishable. The diameter of the glomerulus increases from 72.5 microns in the newborn to 91.7 microns in the adult, an increase of about 26%. The length of the median sagittal section increases from 2.45 mm in the newborn to 7.25 mm in the adult, an increase of approximately 190%, and the length of the medulla increases from 0.7 mm to 3.50 mm, or 400%.

In the newborn rat the kidney is functionally immature as well as histologically immature. The infant rat does not have the ability to concentrate its urine (Falk, 1955), and it is probable that the newborn mouse kidney is functionally immature also. Physiological studies might be done to determine (1) if the infant mouse is unable to regulate the volume of its urine, (2) if so, the age at which the infant mouse kidney develops concentrating ability, and (3) the factors involved in the delay of this function.

The concentrating mechanism depends on countercurrent exchangers or multipliers, which set up an increasing gradient of osmolality in the loops of Henle and vasa recta (Gottschalk and Mylle, 1959). The loops of Henle of the juxtamedullary nephrons dip far down into the medullary pyramids, then turn back toward the cortex, the ascending limb running parallel to the descending limb (Berliner, 1961). Sodium ion is pumped out of the ascending limb into the interstitial fluid, which causes water to move out of descending limb osmotically, concentrating the urine. It is probable that the late development of the medulla and renal papilla with its loops of Henle is responsible for the delay in concentrating ability. The ratio of length of medulla to length of kidney more than doubles between birth and 14 days (Table 1). It is very likely that concentrating ability develops within this period.

Table 1. Relationship of length of medulla to length of kidney. (Ranges indicated in parenthesis)

Age	Length of Medulla*	Length of Kidney**	Ratio of Medulla Length to Kidney Length
Newborn	0.70mm(±.03)	2.45mm(±.01)	0.29
7 day	1.93mm(±.03)	4.25mm(±.03)	0.45
14 day	3.38mm(±.05)	5.63mm(±.03)	0.60
17 day	3.65mm(±.03)	5.68mm(±.07)	0.64

*Measured from the junction with the cortex to the tip of papilla.

**Length of median sagittal section.

SUMMARY

A postnatal histological study of the development of the albino mouse kidney from the newborn to the 22 day old reveals the following maturation changes. Many developing renal corpuscles can be seen in the outer cortex during the first week of life. At seven days of age the proximal tubules have brush borders and basal striations. The papilla is very short in the newborn but continues to increase in length for 17 days. The epithelium of the papillary ducts changes from tall columnar in the newborn to high cuboidal in the 17 day old mouse. At birth large areas of undifferentiated tissue are present in the outer medulla and persist in decreasing amounts through the sixteenth day after birth. The undifferentiated mesenchyme changes first into bipolar cells resembling fibroblasts, then epithelioid cells and finally into primitive tubules. At 17 days all undifferentiated tissue has disappeared and the tubules can be clearly identified. The 17 day old kidney is histologically mature. The ratio of length of medulla to length of kidney more than doubles between birth and 14 days. This study points out the need for physiological studies to determine the ability of the mouse kidney to concentrate urine at various stages of development.

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