A FURTHER REPORT ON THE PRODUCTION OF EXPERI-
MENTAL CHRONIC NEPHRITIS IN ANIMALS
BY THE ADMINISTRATION OF
URANIUM NITRATE *

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In a previous report¹ I was able to show that by the administration of uranium nitrate to guinea-pigs it is possible to produce in them a condition which is analogous to various stages of chronic diffuse nephritis in man. These changes were quite constantly produced, and were found after the administration of uranium nitrate in any one of the following three ways: first, after a series of frequently repeated injections of very small doses of the drug; second, after a series of injections of moderately large doses at intervals of a considerable length of time; and third, after the injection of a single sublethal dose.

The earlier experiments were performed exclusively on guinea-pigs, but during the past three years a large number of experiments have been performed on guinea-pigs, rabbits and dogs in the hope of confirming my previous observations and of producing the condition among animals of different kinds which would be better suited for secondary experiments. In all of these types of animals it has been possible to produce lesions which are characteristic and which are very similar in practically every case. It is true that the histological picture differs considerably from that which is found in human nephritis, but the gross appearance of the kidney in advanced cases is that of granular atrophy, and the histological picture is that of an extreme and progressive fibrosis with accompanying changes in the tubules and glomeruli, so that one cannot do otherwise than describe the condition as a true chronic diffuse nephritis. It is of interest that these results are seemingly at variance with those obtained by Christian and Smith,² who were unable to reproduce conditions similar to those which I had previously reported in guinea-pigs, and who also found that in rabbits they obtained less satisfactory results with uranium nitrate than with potassium bichromate.

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¹From the Pathological Laboratory of Cooper Medical College and Leland Stanford Jr. University.

²A portion of these experiments were performed under a grant from The Rockefeller Institute for Medical Research.

1. Dickson, E. C.: A Report on the Experimental Production of Chronic Nephritis in Animals by the Use of Uranium Nitrate, *THE ARCHIVES INT. MED.*, 1909, iii, 5 and 6, 375.

2. Smith, R. M.: Experimental Chronic Nephritis. *THE ARCHIVES INT. MED.*, 1911, viii, 481.

Before detailing the protocols of the cases which I wish to report, it would seem advisable to describe briefly the characteristic histological picture of the lesions, since they occur so constantly, not only in the kidneys of the different animals of the same kind and of the same experimental series, but also in the kidneys of animals subjected to different experimental procedures, and, indeed, even in the kidneys of the different kinds of animals.

The kidneys all show proliferation of the interstitial connective tissue, which is constantly much more marked and much more fibrous in the inner zone of the cortex. From this inner fibrotic area rays of more cellular connective tissue radiate out toward the surface, and in the more severe cases which have granular surfaces these rays extend to the surface and correspond to the small depressions on it. In the majority of cases the processes are fairly well restricted to the medullary rays, but in the more severe cases, especially in the kidneys of dogs, there is a marked, diffuse proliferation of connective tissue all through the cortex, so that practically no normal appearing tissue remains. In all cases the connective tissue is quite cellular in the outer zone of the cortex, and in many cases there are large areas of dense round cell infiltration in the region of the larger vessels and around some of the glomeruli. In many cases there is a more or less well marked proliferation of connective tissue in the outer portion of the medulla.

The glomerular changes vary considerably, depending on the situation of the glomeruli in the cortex and the severity of the fibrosis of the kidney; and they are always much more severe in the inner zone of the cortex where the connective tissue proliferation is most marked. In every case the glomerular lesions would seem to be of secondary nature, since in the acute uranium nephritis the glomeruli show practically no lesions.³ The most constant change is that of pyknosis and fragmentation of the nuclei in the tuft. In some more severe cases there seems to be an increase in the connective tissue elements in the tufts, and in a few cases, especially in rabbits, there is distinct thickening of the capillary walls in the tufts, and a deposit of some hyaline substance in some of the tufts. In the areas where there is marked fibrosis, many of the glomeruli are completely collapsed. In practically all cases there is more or less marked thickening of the glomerular capsules, and in many there is proliferation of the capsular epithelium. In many cases there is exudation into the capsular spaces, and in some there is also detached epithelium which seems to be crowded into them from the adjoining tubule. In many cases the capsules are distended and form quite large cysts. In those more

3. Christian has described the occurrence of small, oval or round hyaline bodies in the glomerular tufts of rabbits which were subjected to acute uranium poisoning. *Boston Med. and Surg. Jour.*, 1908, clix, 8; and *THE ARCHIVES INT. MED.*, 1911, viii, 469.

severe cases in which the majority of the glomeruli are extensively damaged, the few more normal appearing glomeruli are much larger than normal, often showing over twice the normal diameter, a condition undoubtedly due to a compensating hypertrophy.

There is very constantly a marked parenchymatous change in the tubular epithelium, although the extent of this depends largely on the time which has elapsed since the last injection of uranium. The convoluted tubules are, as a rule, less severely damaged than are the smaller tubules, but there is usually marked hydropic degeneration of the epithelium, and often a coarse, granular degeneration in which the granules take a deep eosin stain. Occasionally the convoluted tubules show pyknosis and desquamation of the epithelium, and contain much exudate and debris in their dilated lumens. A few tubules are widely dilated and form small cysts. The most severe and most constant parenchymatous degeneration is found in the ascending limbs of Henle, and in the smaller connecting tubules. Here there is marked pyknosis, desquamation, and often complete necrosis of the epithelium, and frequently almost complete blocking with desquamated cells and dense hyaline casts. In the few cases where fatty degeneration is present, it is almost wholly confined to the epithelium of the ascending limbs of Henle. In the areas of fibrosis many of the tubules are collapsed and shrunken, and in many places are represented by small accumulations of pyknotic and fragmented nuclei.

In the majority of cases the collecting tubules are quite normal in appearance excepting for the presence of exudate, desquamated cells and casts in their lumens, but in a few cases there is actual destruction and desquamation of the collecting tubule epithelium, and in some places there seems to be an attempt at regeneration of the epithelial covering.

In none of the cases is there any constant and characteristic change in the smaller blood-vessels. In practically all of the more fibrotic kidneys there is a definite peri-arterial fibrosis which seems to be more marked in dogs than in the other animals, but nowhere could be found changes in the intima and media of the smaller arteries such as are constantly present in chronic diffuse nephritis in man.

A fairly complete review of the literature on experimental chronic nephritis may be found in my previous report,¹ and also in reports made by Ophüls,⁴ Lyon,⁵ Pearce⁶ and Christian,⁷ so that a further review will not be made at this time.

4. Ophüls: Experimental Chronic Nephritis. *Jour. Am. Med. Assn.*, 1907, xlviii, 483.

5. Lyon: Experimental Study on the Action of Some Poisons and Toxins on the Kidney. *Jour. Path. and Bact.*, 1903-04, ix, 400.

6. Pearce: Problems of experimental nephritis. *THE ARCHIVES INT. MED.*, 1910, v, 133.

7. Christian: Experimental Nephritis. *Boston Med. and Surg. Jour.*, 1908, clviii, 416.

The method of experiment was the same as employed in my first series. The uranium nitrate was injected subcutaneously in an aqueous solution, and the number and frequency of the injections was controlled chiefly by the variations in the weight of the animals. Routine urine examinations were not made except in isolated cases, but from time to time specimens of urine from the dogs were collected and subjected to the usual clinical methods of examination.

The histological picture of acute uranium nephritis has been so frequently described that I shall not include in the present series any of the animals which succumbed during the first four or five weeks of the experiment.

A. FREQUENTLY REPEATED INJECTIONS OF SMALL DOSES

I. EXPERIMENTS ON GUINEA-PIGS

Protocol 1.—Guinea-pig 2, 1909, received twenty-six injections of 0.5 mg., and seven injections of 1 mg. of uranium nitrate during 217 days. The animal died after receiving the series of 1 mg. doses. Weight at beginning of experiment 750 gm. and at the time of death 650 gm.

Autopsy: No subcutaneous edema, no ascites; 5 c.c. of urine showed much albumin and sugar and a few granular casts; kidneys normal in size, pale, capsule slightly adherent, cut surface cloudy and pale, medulla darker than cortex; liver somewhat congested, right ventricle dilated, aorta apparently normal.

Microscopical Examination: The connective tissue proliferation is not very marked and is confined to the inner zone of the cortex. There are many areas of round cell infiltration in the cortex. There is very marked parenchymatous change, especially in the smaller tubules, with much pyknosis and desquamation, and many hyaline casts.

Protocol 2.—Guinea-pig 3, 1909, received eighty injections of 0.5 mg., and four injections of 1 mg. of uranium nitrate during 696 days. The animal died two days after the last injection of 1 mg. Weight at beginning of experiment 550 gm., maximum weight 860 gm., weight at time of death 510 gm.

Autopsy: No subcutaneous edema, ascites or hydrothorax; 5 c.c. of urine showed no albumin or casts; kidneys normal size, red, smooth except for a few small dimples which are more numerous on the right kidney; capsule not adherent, cut surface cloudy, markings fairly distinct; liver congested, right ventricle dilated, aorta apparently normal.

Microscopical Examination: Moderate proliferation in the interstitial connective tissue, in the usual location; very little parenchymatous change; glomerular and tubular changes characteristic.

Protocol 3.—Guinea-pig 4, 1909, received seventy injections of 0.5 mg. of uranium nitrate during 590 days, and died on the 595th day. Weight at beginning of experiment 440 gm., maximum weight 790 gm., weight at time of death 720 gm.

Autopsy: No subcutaneous edema or urine; about 30 c.c. of slightly blood-stained fluid removed from peritoneal sac; kidneys small, surface pale with small hemorrhagic spots, capsule not adherent, cut surface somewhat cloudy; intestines much congested, liver pale, heart and aorta apparently normal.

Microscopical Examination: Moderate increase of interstitial connective tissue, almost entirely located in inner zone. Characteristic glomerular and tubular changes in the fibrotic areas. Little parenchymatous change in tubular epithelium. Many hyaline casts.

Protocol 4.—Guinea-pig 5, 1909, received eighty-six injections of 0.5 mg. of uranium nitrate during 695 days, and died six days after the last injection;

weight at the beginning of the experiment 520 gm., maximum weight 800 gm., weight at time of death 500 gm.

Autopsy: No subcutaneous edema, slight ascites and hydrothorax (10 c.c. of fluid collected from peritoneal sac and about 1 c.c. from each pleural sac). A few c.c. of urine showed a trace of albumin but no casts; kidneys about normal size, dark red, capsule definitely adherent, surface everywhere finely granular, cut surface bulges and is cloudy; no cysts seen; lungs and liver congested, right ventricle dilated, aorta apparently normal.

Microscopical Examination: Very marked proliferation of interstitial connective tissue in both inner and outer zones of cortex; many rays extend to the surface; glomeruli show characteristic changes; some are cystic; tubular epithelium shows little degeneration except in fibrotic areas; many hyaline casts.

Protocol 5.—Guinea-pig 6, 1909, received forty-seven injections of 0.5 mg. of uranium nitrate during 360 days, and died on the 434th day. Weight at beginning of experiment 660 gm., maximum weight 770 gm., weight at time of death 640 gm.

Fig. 1.—Guinea-Pig 14, Series B, Protocol 7. $\times 25$. Note the wide distribution of the connective tissue proliferation in the inner zone of the cortex, and the rays extending toward the surface; also the depression at the place where one ray reaches the surface.

Autopsy: No subcutaneous edema and no urine; there was a small amount of fluid in the peritoneal sac and the stomach was distended with fluid; kidneys slightly smaller than normal, mottled, slightly dimpled, capsule not adherent, cut surface cloudy, not swollen; liver congested, heart and aorta apparently normal.

Microscopical Examination: Slight connective tissue proliferation in inner zone of cortex; some glomeruli show characteristic changes and a few are cystic; very marked parenchymatous change with much desquamation; many hyaline casts.

Protocol 6.—Guinea-pig 27, 1909, received thirty-seven injections of 0.5 mg. of uranium nitrate during 360 days, and died on the 429th day. Weight at beginning of experiment 570 gm., maximum weight 880 gm., weight at time of death 630 gm.

Autopsy: No subcutaneous edema and no urine; a small amount of fluid in peritoneal sac; kidneys normal size, capsule slightly adherent, surface dark red and finely granular, cut surface not cloudy, markings fairly distinct; liver and lungs congested, aorta apparently normal; right ventricle somewhat dilated.

Microscopical Examination: Moderate proliferation of interstitial connective tissue with many rays extending to the surface; glomeruli show usual changes, and some are cystic; there is moderate parenchymatous change with desquamation in the smaller tubules, and many hyaline casts; the tubules in the fibrotic areas are shrunken and collapsed.

Fig. 2.—Guinea-Pig 14. $\times 90$. Note the proximity to a large blood vessel, the marked fibrosis, and the shrinkage of tubules in the fibrotic area.

II. EXPERIMENTS ON RABBITS

Protocol 1.—Rabbit 11, 1909, received thirty-two injections of 1 mg. of uranium nitrate during 196 days, and died on the 203d day. Weight at beginning of experiment 3.075 gm. and at the time of death 2.020 gm.

Autopsy: No subcutaneous edema, no ascites, and no urine. Double fibrino-purulent pleurisy. Kidneys somewhat enlarged, surface smooth, capsule not adherent, cut surface bulged and distinctly cloudy. Liver congested, right ventricle dilated, aorta apparently normal.

Microscopical Examination: Marked diffuse proliferation of interstitial connective tissue, with many rays extending to the surface, and with many areas of round cell infiltration; glomeruli show thickening of capillary walls, and much

exudate in capsular spaces; marked parenchymatous degeneration of tubular epithelium, with much desquamation in Henle's loops and collecting tubules, and many hyaline casts; marked atrophy and collapse of tubules in the fibrotic areas.

Protocol 2.—Rabbit 14, 1909, received thirty subcutaneous injections of 1 mg. of uranium nitrate during 193 days, and died on the 196th day. Weight at beginning of experiment 3,250 gm., at time of death 2,430 gm.

Autopsy: No subcutaneous edema, no ascites, no urine; kidneys large, pale, smooth, with numerous small scars on surface, capsule not adherent; on section was some bulging and marked opacity; the cortex was pale and the medulla dark red; liver congested, right ventricle dilated, aorta apparently normal; marked double fibrino-purulent pleurisy.

Fig. 3.—Guinea-Pig 8, Series B, Protocol 1. $\times 100$. Note the marked periarterial fibrosis, and the proximity of the damaged glomeruli to the vessel.

Microscopical Examination: Very marked, diffuse proliferation of interstitial connective tissue, with many large areas of round cell infiltration. The majority of the glomeruli and tubules are damaged; marked parenchymatous degeneration; much desquamation in smaller tubules and many hyaline casts.

Protocol 3.—Rabbit 15, 1909, received thirty-three subcutaneous injections of 1 mg. of uranium nitrate during 274 days, and was killed on the 457th day. Weight at the beginning of the experiment 2,850 gm., at the time of death 2,950 gm.

Autopsy: No subcutaneous edema, slight ascites, no urine; kidneys rather small, pale and slightly granular; capsule not adherent; on the surface

were a few small hemorrhagic spots and a few small cysts. On section the cut surface was pale and the cortex was apparently thinner than normal.

Microscopical Examination: Very marked proliferation of interstitial connective tissue, with marked inner fibrosis, and many rays extending to surface; marked glomerular changes; capillary walls of tufts thickened, and many of the capsules cystic; moderate parenchymatous change, little desquamation, many tubules involved in fibrotic areas, shrunken; many hyaline casts.

Protocol 4.—Rabbit 18, 1909, received thirty-four injections of 1 mg. of uranium nitrate during 278 days, and died about thirteen months after the first injection.

Autopsy: Kidneys rather small, pale, finely granular, capsule not adherent; cut surface not swollen, cortex thinner than normal.

Microscopical Examination: Very marked and very diffuse proliferation of interstitial connective tissue, with many rays of cellular connective tissue extending outwards from the inner fibrotic area; very many areas of round cell infiltra-

Fig. 4.—Rabbit 41, Series B, Protocol 12. $\times 40$. Note the wide distribution of the lesion, the greater severity in the inner zone of the cortex, and the large glomerular cyst.

tion in the cortex; extensive glomerular changes; thickening of capillary walls, increase in connective tissue in tufts, and dilatation of capsular spaces; marked parenchymatous degeneration of tubules with much shrinkage and desquamation; marked desquamation of epithelium of collecting tubules; many hyaline casts; marked atrophy and collapse of tubules in the fibrotic areas.

B. OCCASIONAL SUBLETHAL DOSES

1. EXPERIMENTS ON GUINEA-PIGS

Protocol 1.—Guinea-pig 8, 1909, received ten injections of 5 mg. of uranium nitrate during 461 days, and died two days after the last injection; weight at beginning of experiment 620 gm., maximum weight 690 gm., weight at time of death 600 gm.

Autopsy: Very marked subcutaneous edema over belly and legs; no ascites, hydrothorax or urine; kidneys very small, about two-thirds normal size, surface pale, granular, capsule adherent, cut surface pale and cloudy, cortex thin; no cysts seen; liver and lungs congested, heart large, right ventricle dilated, aorta apparently normal.

Microscopical Examination: Marked proliferation of interstitial connective tissue with many rays extending to the surface; little round cell infiltration; moderate characteristic changes in the glomeruli and tubules in the fibrotic areas; some glomerular cysts; relatively little parenchymatous change in the more normal appearing tubules; many hyaline casts.

Protocol 2.—Guinea-pig 9, 1909, received six injections of 5 mg. of uranium nitrate during 110 days, and died seven days after the last injection; weight at beginning of experiment 725 gm., at time of death 435 gm.

Autopsy: No subcutaneous edema, no ascites, no urine; kidneys slightly swollen, pale, slightly dimpled, capsule not adherent; cut surface cloudy and

Fig. 5.—Rabbit 41. $\times 100$. Note the diffuse distribution of the connective tissue proliferation, the collapse of the tubules and the pyknosis and desquamation of the tubular epithelium.

swollen, cortex paler than medulla; liver congested, right ventricle dilated; aorta apparently normal.

Microscopical Examination: Very marked diffuse proliferation of interstitial connective tissue which is very cellular; little shrinkage; marked glomerular changes with many cysts; marked parenchymatous change with much desquamation and many dense hyaline casts.

Protocol 3.—Guinea-pig 10, 1909, received eight injections of 5 mg. of uranium nitrate during 400 days, and died five days after the last injection; weight at beginning of experiment 520 gm., maximum weight 830 gm., weight at time of death 470 gm.

Autopsy: Moderate subcutaneous edema over belly, thorax and legs; 30 c.c. fluid removed from peritoneal sac, and several c.c. from both pleural sacs; peri-

cardium was distended with fluid to about twice normal size; no urine; kidneys about normal size, dark red, slightly dimpled, capsule slightly adherent, cut surface cloudy, not swollen; no cysts seen; liver congested, right ventricle dilated, aorta apparently normal.

Microscopical Examination: Slight diffuse proliferation of interstitial connective tissue and many areas of round cell infiltration; relatively little glomerular change although a few capsules are cystic; very marked parenchymatous degeneration in all tubules, much desquamation and many hyaline casts.

Protocol 4.—Guinea-pig 11, 1909, received four injections of 5 mg. of uranium nitrate during 248 days, and died eleven days after the last injection: weight

Fig. 6.—Rabbit 50, Series B, Protocol 6. $\times 90$. Note the dense fibrosis, the fibrous thickening of the glomerular capsules and the marked destruction of tubules in the fibrous area.

at beginning of experiment 590 gm., at time of last injection 730 gm., and at time of death 855 gm., an increase of 125 gm. during the last eleven days of life.

Autopsy: Very extreme subcutaneous edema over belly, thorax and legs; very marked ascites (110 c.c. collected and at least 30 c.c. escaped), a few drops of fluid in both pleural sacs, and moderate distention of pericardium; no urine; kidneys swollen, surface mottled, smooth, capsule not adherent, cut surface bulging, edematous and cloudy; liver congested and containing small focal necroses; moderate perihepatitis and perisplenitis; spleen large, right ventricle markedly dilated, aorta apparently normal.

Microscopical Examination: Some cellular proliferation of interstitial connective tissue, and a few areas of round cell infiltration; relatively little glomerular change; very marked parenchymatous change, much desquamation and a few hyaline casts.

Protocol 5.—Guinea-pig 12, 1909, received four injections of 5 gm. of uranium nitrate during seventy-five days, and died nine days after the last injection; weight at beginning of experiment 410 gm., maximum weight 480 gm., weight at time of death 295 gm.

Autopsy: No subcutaneous edema, no ascites, no urine; kidneys normal size, surface smooth with small dimples and a few pin point hemorrhages, capsule not

Fig. 7.—Rabbit 106, Series B, Protocol 11. $\times 90$. Note the marked increase in fibrous tissue, the dense round cell infiltration, the thickening of the glomerular capsules, and the atrophy of the tubules.

adherent; cut surface swollen and cloudy; liver congested, right ventricle dilated, aorta apparently normal.

Microscopical Examination: Slight cellular proliferation of connective tissue in inner zone of cortex and a few areas of round cell infiltration around some of the glomeruli; very marked parenchymatous change affecting all tubules, very numerous hyaline casts and a few cystic glomeruli.

Protocol 6.—Guinea-pig 13, 1909, received four injections of 5 mg. of uranium nitrate during seventy-seven days, and died three days after the last injection; weight at beginning of experiment 540 gm., weight at time of death 500 gm.

Autopsy: No subcutaneous edema, no ascites, no urine; kidneys normal size, surface shows few small dimples, capsule not adherent; cut section cloudy; a few small cysts; liver congested, right ventricle dilated, aorta apparently normal.

Microscopical Examination: Slight cellular infiltration in inner zone of cortex with some areas of round cell infiltration in outer zone; little glomerular and tubular change; a few glomerular cysts and a few hyaline casts.

Protocol 7.—Guinea-pig 14, 1909, received seven injections of 5 mg. of uranium nitrate during 400 days, and died seven days after the last injection; weight at beginning of experiment 550 gm., maximum weight 680 gm., weight at time of death 310 gm.

Autopsy: No subcutaneous edema or hydrothorax; a few c.c. of ascitic fluid removed from peritoneal sac; 5 c.c. of urine showed a trace of albumin but no casts; kidneys small, pale, fibrous in appearance, surface granular, capsule adherent, cortex thin, cut surface slightly opaque. Liver and lungs congested, left ventricle seems thicker than normal, aorta apparently normal.

Microscopical Examination: Very marked diffuse proliferation of interstitial connective tissue and many large areas of round cell infiltration; very marked glomerular and tubular destruction in the fibrotic areas; little parenchymatous change in the more normal appearing tubules; many dense hyaline casts.

Fig. 8.—Dog 6, Series 1909, Protocol 5. Actual size. Note the marked dimpling of the surface, and the tears where the capsule was adherent.

Protocol 8.—Guinea-pig 17, 1909, received two injections of 10 mg. of uranium nitrate, with an interval of seventy-seven days, and died seven days after the last injection; weight at beginning of experiment 570 gm., weight ten days before death 650 gm., weight at time of death 380 gm.

Autopsy: No subcutaneous edema, no ascites, no urine; kidneys swollen, mottled, cloudy and smooth; capsule not adherent; liver congested, right ventricle dilated, aorta apparently normal.

Microscopical Examination: Fairly diffuse cellular proliferation of interstitial connective tissue in inner zone of cortex; very marked parenchymatous degeneration with much desquamation and many hyaline casts; a few glomeruli contain exudate in their capsular spaces and some are dilated into small cysts.

Protocol 9.—Guinea-pig 18, 1909, received three injections, respectively, of 15, 10 and 5 mg. of uranium nitrate during 485 days, and died eleven days after the last injection; weight at beginning of experiment 800 gm., maximum weight 910 gm.; weight at time of death 540 gm.

Autopsy: No subcutaneous edema, 10 c.c. ascitic fluid and about 1 c.c. of fluid in each pleural sac and in pericardial sac; kidneys small, pale, granular with

adherent capsule, cortex thin, cut surface cloudy, no cysts seen: 10 c.c. urine showed heavy cloud of albumin but no casts.

Microscopical Examination: Moderate diffuse proliferation of interstitial cellular connective tissue, many areas of round cell infiltration and moderate fibrosis around the larger blood vessels; moderate glomerular change with many large cysts; moderate parenchymatous change, especially in the smaller tubules; many hyaline casts.

Protocol 10.—Guinea-pig 19, 1909, received two injections of 10 mg. of uranium nitrate with an interval of seventy-seven days, and died five days after the second injection.

Autopsy: No subcutaneous edema, ascites, or urine; kidneys mottled, smooth, with a few dimples, capsule not adherent, cortex cloudy; liver congested, heart and aorta apparently normal.

Microscopical Examination: Practically the same as described in Protocol 8.

Fig. 9. — Dog 3, Series 1910, Protocol 8. $\times 25$. Note the wide distribution and the severity of the lesions, the greater amount of connective tissue proliferation in the inner zone of the cortex, and the radiating rays extending toward the surface.

Protocol 11.—Guinea-pig 20, 1909, received three injections, respectively, of 10, 5 and 5 mg. of uranium nitrate during 485 days, and died five days after the last injection. Weight at beginning of experiment 610 gm., maximum weight 750 gm., weight at time of death 505 gm.

Autopsy: Moderate subcutaneous edema, moderate ascites and a few c.c. of urine which showed definite albumin and many granular casts; kidneys large, swollen, congested, capsule not adherent, cut surface swollen and cloudy; liver and lungs congested, right ventricle dilated, aorta normal.

Microscopical Examination: Irregularly scattered areas of fibrosis and diffuse proliferation of interstitial cellular connective tissue in cortex; marked cystic

dilatation of both tubules and glomeruli; the majority of the glomeruli show little damage; relatively little parenchymatous change; many hyaline casts.

B. EXPERIMENTS ON RABBITS

Protocol 1.—Rabbit 23, 1909, received 5 injections of 5 mg. of uranium nitrate during 111 days, and died on the 175th day.

Autopsy: No subcutaneous edema or ascites. Fifty c.c. of urine showed very marked albumin and many hyaline and granular casts. Kidneys swollen, dark red, surface smooth, capsule not adherent, cut surface cloudy. Liver congested, heart and aorta apparently normal.

Microscopical Examination: Marked proliferation of interstitial connective tissue. Marked glomerular change, with a hyaline deposit in some of the tufts. Many hypertrophied glomeruli. Marked destruction of tubules in fibrotic areas; relatively little parenchymatous change in more normal appearing tubules. Many hyaline casts.

Fig. 10.—Dog 3, Series 1910. $\times 100$. Note the extreme fibrosis and round cell infiltration, the thickening and beginning cyst formation of the glomerular capsules, and the marked tubular destruction.

Protocol 2.—Rabbit 24, 1909, received four injections of 5 mg. of uranium nitrate during eighty-one days, and died seven days after the last injection.

Autopsy: No subcutaneous edema and no ascites; 10 c.c. of urine showed a moderate cloud of albumin and many granular casts; kidneys large, surface shows many small scars and some small hemorrhages, capsule slightly adherent, cut surface cloudy and swollen; liver congested, left ventricle seemed thicker than normal, aorta apparently normal.

Microscopical Examination: Very marked diffuse proliferation of interstitial connective tissue involving the whole cortex and outer part of the medulla; little fibrosis and little shrinkage; moderate glomerular change with some fibrosis and some hyaline deposit in the tufts; many of the tubules are separated by cellular proliferation of connective tissue; marked parenchymatous change, some desquamation, and many hyaline casts.

Protocol 3.—Rabbit 25, 1909, received three injections of 5 mg. of uranium nitrate during seventy-four days, and was killed on the 281st day; weight at beginning of experiment 2,200 gm., at the time of death 2,930 gm.

Autopsy: No subcutaneous edema or ascites; urine showed no albumin or casts; kidneys normal size, congested, capsule not adherent, surface smooth with few small scars, cut surface not swollen or cloudy; heart, liver and aorta normal.

Microscopical Examination: Scattered areas of cellular connective tissue proliferation in cortex and a few small areas of fibrosis in inner zone; majority of glomeruli and tubules normal; a few glomeruli have thickened capsules and pyknosis and fragmentation of nuclei, and a few tubules show hydropic and some granular degeneration.

Fig. 11.—Dog 5, Series 1909, Protocol 4. $\times 90$. Note the extreme degree of connective tissue proliferation, the cellular character of the newly-formed connective tissue, the marked shrinkage of the glomeruli, and the extensive tubular destruction.

Protocol 4.—Rabbit 26, 1909, received two injections of 5 mg. of uranium nitrate, with an interval of thirty-two days, and died seven days after the second injection.

Autopsy: No subcutaneous edema and no ascites; 25 c.c. urine showed a trace of albumin and many hyaline and epithelial casts; kidneys swollen, congested, surface smooth and mottled, capsule not adherent, cut section swollen and opaque; liver large and congested; heart and aorta normal.

Microscopical Examination: Very marked, diffuse, cellular proliferation of interstitial connective tissue in outer and inner zone of cortex; little fibrosis; many large areas of round cell infiltration in cortex and medulla; moderate glomerular and tubular change in fibrotic areas and marked parenchymatous degeneration with some desquamation and many hyaline casts.

Protocol 5.—Rabbit 27, 1909, received three injections of 5 mg. of uranium nitrate during seventy-four days and was killed on the 112th day. There had been a large abscess on the shoulder for about three weeks previous to death.

Autopsy: No subcutaneous edema, slight ascites and 30 c.c. of urine, which shewed a trace of albumin but no casts; kidneys large, pale, smooth with a few dimples, capsule not adherent, cut surface swollen, cloudy and edematous; liver congested, heart and aorta apparently normal.

Fig. 12.—Dog 6, Series 1909, Protocol 5. $\times 60$. Note the very marked peri-arterial fibrosis.

Microscopical Examination: Marked proliferation of interstitial connective tissue, with many areas of round cell infiltration in cortex; very marked glomerular and tubular change, especially in inner zone of cortex; many large glomerular cysts; marked parenchymatous change in tubules; much desquamation and many hyaline casts.

Protocol 6.—Rabbit 50, 1910, received one injection of 5 mg., and two injections of 3 mg. of uranium nitrate during 158 days, and died on the 264th day. Weight at beginning of experiment 1.680 gm., maximum weight 2.460 gm., weight at time of death 1.175 gm.

Autopsy: No subcutaneous edema, a few c.c. of ascitic fluid, and 15 c.c. urine, which showed a trace of albumin, many hyaline and waxy casts, and a few granular casts; kidneys small, pale, capsule not adherent, surface finely granular, cut section opaque, cortex thin, medulla edematous; liver congested, right ventricle dilated, left ventricle distinctly thicker than normal, aorta apparently normal.

Microscopical Examination: Marked diffuse proliferation of interstitial connective tissue. Marked glomerular change in inner zone of cortex, and some compensatory hypertrophy of glomeruli in outer zone; moderate parenchymatous change, some desquamation, and a few hyaline casts.

Protocol 7.—Rabbit 61, 1910, received three injections of 3 mg. of uranium nitrate during 150 days, and died on the 190th day. Weight at beginning of experiment 1,680 gm., maximum weight 1,810 gm., weight at time of death 1,390 gm.

Autopsy: No subcutaneous edema and no ascites; 10 c.c. of urine showed a faint trace of albumin but no casts; kidneys small, pale, finely granular, capsule not adherent, cut surface opaque; lungs and liver congested; heart dilated, aorta apparently normal.

Microscopical Examination: Moderate diffuse proliferation of interstitial connective tissue; relatively little glomerular change; marked parenchymatous change, especially in smaller tubules; many hyaline casts.

Protocol 8.—Guinea-pig 63, 1910, received three injections of 3 mg., and one injection of 2 mg. of uranium nitrate during 202 days, and died on the 242d day. Weight at beginning of experiment 1,730 gm., maximum weight 2,210 gm., weight at time of death 1,860 gm.

Autopsy: No subcutaneous edema, marked adhesive peritonitis with old adhesions between all viscera; 10 c.c. of urine showed definite cloud of albumin but no casts; kidneys normal size, pale, mottled, finely granular, capsule not adherent, cut surface opaque; liver and lungs congested, right ventricle widely dilated, left ventricle in extreme systole, hard, and apparently definitely hypertrophied, aorta normal.

Microscopical Examination: Moderate diffuse proliferation of interstitial connective tissue, with many areas of round cell infiltration scattered irregularly through the cortex; moderate glomerular change, with some compensatory hypertrophy of glomeruli in the outer zone; moderate parenchymatous change, and many dense hyaline casts.

Protocol 9.—Rabbit 67, 1910, received three injections of 3 mg. of uranium nitrate during 150 days, and died on the 175th day; weight at beginning of experiment 1,720 gm., maximum weight 2,010 gm., weight at time of death 1,250 gm.

Autopsy: No subcutaneous edema, ascites or hydrothorax; 20 c.c. urine showed a trace of albumin but no casts; kidneys small, slightly congested, surface smooth with a few dimples, capsule not adherent, cut surface cloudy, some edema in pyramids; liver and lungs congested, right ventricle dilated, aorta apparently normal.

Microscopical Examination: Moderate diffuse proliferation of interstitial connective tissue which is much more cellular than usual; moderate glomerular change especially in inner zone; moderate parenchymatous change with desquamation, and many hyaline casts.

Protocol 10.—Rabbit 68, 1910, received three injections of 3 mg. of uranium nitrate during 150 days, and died six days after the last injection; weight at beginning of experiment 1,580 gm., maximum weight 1,780 gm., weight at time of death 1,200 gm.

Autopsy: No subcutaneous edema or ascites; a few c.c. of urine showed a marked cloud of albumin, many hyaline and a few granular casts; kidneys about normal size, capsule not adherent, surface dark red in color and everywhere covered by many large scars, cut surface cloudy.

Microscopical Examination: Very marked diffuse proliferation of interstitial connective tissue, and many areas of round cell infiltration in cortex; fairly

marked glomerular changes; marked parenchymatous degeneration of tubular epithelium; many hyaline casts.

Protocol 11.—Rabbit 106, 1910, received two injections of 3 mg. of uranium nitrate with an interval of 105 days, and died on the 127th day. Weight at beginning of experiment 2,260 gm., at time of death 1,200 gm.

Autopsy: No subcutaneous edema or ascites; right sided pleuropneumonia. A few c.c. of urine showed a trace of albumin and some hyaline casts; kidneys small, capsule adherent, surface pale and finely granular, cut surface pale, cortex thin; liver congested, right ventricle dilated, aorta apparently normal.

Microscopical Examination: Very marked diffuse proliferation of interstitial connective tissue with many rays extending to surface, and much diffuse proliferation of connective tissue between the rays; moderate glomerular change, and marked parenchymatous degeneration of tubular epithelium with some desquamation and many hyaline casts.

Protocol 12.—Rabbit 41, 1910, received five injections of 3 mg. and one injection of 6 mg. of uranium nitrate during seventy-two and one-half weeks, and died thirty-six days after the last injection. Weight at beginning of experiment 1,660 gm., maximum weight 2,720 gm., weight at time of last injection 2,550 gm., and weight at time of death 1,770 gm.

Autopsy: No subcutaneous edema; moderate ascites, hydrothorax and hydropericardium; 20 c.c. urine showed much albumin and a few hyaline casts; kidneys large, mottled, capsule not adherent, surface finely granular, cut surface pale, cortex distinctly narrow, much edema in medulla; liver large, firm, rough with evidence of passive congestion and increase in connective tissue; heart large, left ventricle evidently hypertrophied, right ventricle dilated; lungs congested, adrenals large, spleen and aorta apparently normal; chronic adhesive peritonitis.

Microscopical Examination: Very marked diffuse proliferation of interstitial connective tissue with many rays extending to the surface, much proliferation in areas of cortex between the rays, and much round cell infiltration; moderate glomerular change, much pyknosis and fragmentation of nuclei of tufts, little thickening of capsule or proliferation of capsular epithelium; some exudate into capsular spaces and moderate cyst formation; very marked parenchymatous degeneration with complete necrosis in many tubules and much desquamation; evidence of epithelial regeneration in many tubules; few hyaline casts; marked periarterial fibrosis around many of medium sized and larger arteries.

EXPERIMENTS ON DOGS

The injections on dogs during the earlier experiments were made somewhat at random in order to ascertain the degree of tolerance of these animals for the drug, as well as the effects which would be produced on the kidneys. At times the injections were repeated at frequent, and at other times, at much longer intervals. It was found that the best results were obtained either when fairly large amounts of the drug were injected during as short a period of time as the animal could stand, or, when almost sublethal doses were given, at longer intervals. The maximum dose which can be used with safety is approximately 3 mg. per kilogram of weight. This amount produces a fairly acute attack of nephritis from which the animal usually recovers, and can often be safely repeated several times at intervals of several weeks. In every case, however, the initial dose should be somewhat smaller than this, and subsequent treatment should be based on the reaction which the animal shows to the first injection.

Protocol 1.—Dog 2, 1909, a female fox terrier about 5 months old, received two injections of 5 mg. and one injection of 10 mg. of uranium nitrate during fifty days, and was killed thirty-three weeks after the first injection. The weight at time of death was 5 kilos. The urine several times showed albumin and some casts.

Autopsy: No edema, no ascites; urine free from albumin or casts; kidneys small, slightly mottled, capsules slightly adherent, cut surface opaque but not swollen; heart, liver, lungs and aorta apparently normal.

Microscopical Examination: Little increase in interstitial connective tissue except around the larger blood vessels; several large areas of round cell infiltration in cortex around blood vessels and glomeruli; some glomerular change in inner zone of cortex; some parenchymatous change, more marked in tubules in region of round cell infiltration; no casts seen.

Protocol 2.—Dog 3, 1909, a mongrel male about 5 months old, received two injections of 5 mg. and one of 10 mg. of uranium nitrate during fifty days, and was killed thirty-four weeks after the first injection. Albumin and a few hyaline casts were found once during the course of the experiment. Weight at time of death 10.26 kilos.

Autopsy: No edema and no ascites; urine showed many hyaline and granular casts, but no albumin; kidneys about normal size, slightly mottled, capsule slightly adherent, cut surface cloudy; liver, lungs and aorta apparently normal; heart large and flabby, weight 80 gm., and the heart to body ratio 0.00779.

Microscopical Examination: No diffuse proliferation of interstitial connective tissue, but many foci of cellular proliferation are scattered irregularly through the cortex; some glomerular change, especially near the areas of newly-formed connective tissue. In the inner zone a few glomeruli show almost complete fibrosis of the tufts, and some are cystic; relatively little parenchymatous change; some exudate and desquamation and a few hyaline casts.

Protocol 3.—Dog 4, 1909, a mongrel male about 5 months old, received one injection of 10 mg., and one of 20 mg. of uranium nitrate within seventeen days, and was killed thirty-one weeks after the first injection. Weight at time of death 14.28 kilos. The urine showed albumin and casts several times during the course of the experiment.

Autopsy: No edema or ascites; urine showed a few hyaline casts but no albumin; kidneys slightly larger than normal, mottled, capsule not adherent, surface slightly dimpled, cut surface cloudy and edematous, pyramids more congested than the cortex; liver and lungs congested, aorta apparently normal; heart large, left ventricle apparently hypertrophied, right ventricle dilated, weight 118 gm., and heart to body ratio 0.00802.

Microscopical Examination: Little new formation of fibrous tissue except around the larger blood vessels; several large areas of cellular infiltration in cortex where round cells and fibroblasts widely separate the kidney tubules; little glomerular change except in the areas of cellular proliferation; very marked parenchymatous change in practically all tubules, much pyknosis and desquamation in the smaller tubules, and many dense, hyaline casts.

Protocol 4.—Dog 5, 1909, a mongrel male about 5 months old, received two injections of 10 mg. of uranium nitrate during seventeen days, and was found dead fourteen weeks after the first injection. Weight at time of death 5 kilos.

Autopsy: No edema, ascites or urine; kidneys small, pale with minute hemorrhages, capsule thick and firmly adherent, surface torn and rough, with numerous large and smaller depressions; no cysts seen; cut surface white and fibrous, markings almost obliterated, cortex very narrow; liver large and congested; heart large, left ventricle firm and appears hypertrophied; aorta apparently normal; weight of heart not obtained.

Microscopical Examination: Very marked proliferation of interstitial connective tissue in both cortex and medulla, and marked fibrous thickening of the capsule; newly-formed connective tissue very fibrous in inner zone of cortex

from which more cellular rays extend to surface; marked, diffuse connective tissue proliferation in areas of cortex between the rays; practically all glomeruli extensively involved; much fibrosis of tufts and thickening of capsules, but no cyst formation. A few glomeruli show compensatory hypertrophy; very extensive tubular destruction with much desquamation in smaller tubules, and extreme atrophy of tubules in the fibrotic areas; much more fibrosis in medulla than usual; no casts seen.

Protocol 5.—Dog 6, 1909, a mongrel male about 5 months old, received two injections of 10 mg. of uranium nitrate during seventeen days, and was killed twenty-nine weeks after the first injection. Albumin but no casts were found twice during life. Weight at time of death 5.61 kilos.

Autopsy: No subcutaneous edema and no ascites; 30 c.c. urine showed many granular and hyaline casts but no albumin; kidneys small, pale, capsules firmly adherent, surface torn, finely granular, with some large scars and a few small cysts; cut surface very pale and fibrous, cortex thin, medulla white and glistening; liver large and congested; aorta apparently normal; heart large, left ventricle thick and firm, right ventricle dilated, weight 60 gm.; heart to body ratio 0.01069.

Microscopical Examination: Very marked diffuse proliferation of interstitial connective tissue, more marked and more fibrous in inner zone of cortex, with more cellular rays extending to the surface; much diffuse proliferation of connective tissue in cortex between the rays, and many areas of round cell infiltration; very marked change in practically all glomeruli, much shrinkage and capsular thickening, and many small cysts; some glomeruli show compensatory hypertrophy; practically none of the tubules normal, much dilatation of lumens, little desquamation of epithelium but much exudate; much increase in fibrous tissue in medulla; very few casts seen.

Protocol 6.—Dog 1, 1910, a mongrel female about 9 months old, received four injections of 10 mg. and three injections of 20 mg. of uranium nitrate during forty-five weeks, and was accidentally killed sixty-one weeks after the first injection. Weight at beginning of experiment 7.5 kilos, weight at time of death 7.68 kilos.

Autopsy: No subcutaneous edema or ascites, no urine; kidneys small, reddish brown in color, capsule slightly adherent, surface slightly granular, cut surface apparently normal; liver, lungs and aorta normal; heart rather large, in diastole, weight 61.5 gm.; heart to body ratio 0.00800.

Microscopical Examination: Marked proliferation of interstitial connective tissue, quite cellular in outer zone; much round cell infiltration; moderate glomerular change; marked parenchymatous change; much fat, especially in ascending limbs of Henle; little desquamation; much shrinkage in areas of fibrosis; few hyaline casts.

Protocol 7.—Dog 2, 1910, a male spaniel, about 6 months old, received three injections of 5 mg., one injection of 10 mg., and two injections of 20 mg. of uranium nitrate during fifty-three weeks, and was killed seventy-one weeks after the first injection. Weight at beginning of experiment 5.25 kilos, weight at time of death 16.3 kilos.

Autopsy: No subcutaneous edema, no ascites, no urine; kidneys about normal size, dark red, capsule slightly adherent, surface smooth, cut surface opaque and slightly edematous; liver, lungs, adrenals and aorta apparently normal; heart large, right side dilated, weight 145 gm., heart to body ratio 0.00888.

Microscopical Examination: Marked diffuse proliferation of interstitial connective tissue which is quite cellular, and many large areas of round cell infiltration; marked change in many of the glomeruli, with lobulation and shrinkage of tufts and thickening of capsules; in some glomerular spaces there are masses of epithelium which are evidently pushed in from the tubules; a few glomeruli show compensatory hypertrophy; very marked parenchymatous change in all tubules, marked desquamation in smaller tubules, a few casts.

Protocol 8.—Dog 3, 1910, a mongrel male, about 9 months old, received two injections of 10 mg., two of 30 mg., and three of 50 mg. of uranium nitrate during fifty-three weeks, and was killed sixty weeks after the first injection; weight at beginning of experiment 14.2 kilos, weight at time of death 21.6 kilos; urine examinations during life showed traces of albumin and a few hyaline casts.

Autopsy: No subcutaneous edema, no ascites, no urine; kidneys normal in size, brownish in color, capsule slightly adherent, surface fairly smooth, cut surface cloudy, cortex slightly narrower than normal; liver, pancreas and aorta apparently normal; heart large, left ventricle firm and thick, weight 180 gm.; heart to body ratio 0.00818.

Microscopical Examination: Very marked diffuse proliferation of interstitial connective tissue which in most places does not reach the surface; many areas of round cell infiltration; marked glomerular change, many tufts shrunken, distinct fibrosis of some tufts, much exudate in spaces and marked thickening of capsules; many glomeruli are hypertrophied; very marked parenchymatous change in all tubules, much desquamation, much fatty degeneration in convoluted tubules and ascending limbs of Henle; no casts seen.

Protocol 9.—Dog 5, 1910, a black and tan male about 9 months old, received three injections of 10 mg. and one of 30 mg. of uranium nitrate during twenty-three weeks, and died eleven days after the last injection. Weight at beginning of experiment 8.98 kilos, and at time of death 8.02 kilos.

Autopsy: No subcutaneous edema and no ascites; 30 c.c. of urine in bladder showed much albumin but no casts; kidneys firm, swollen, congested, capsule slightly adherent, surface smooth, cut surface swollen and cloudy; liver, lungs and aorta apparently normal; heart large, right ventricle dilated, left ventricle apparently enlarged, weight 73 gm.

Microscopical Examination: Moderate increase in interstitial connective tissue, many areas of round cell infiltration; very little glomerular change; marked parenchymatous change with some desquamation and many hyaline casts.

Protocol 10.—Dog 7, 1910, a mongrel male about 8 months old, received three injections of 10 mg., two of 30 mg. and three of 40 mg. of uranium nitrate during fifty-three weeks, and died thirteen days after the last injection; weight at beginning of experiment 8.9 kilos, weight at time of last injection 12.9 kilos, and weight at time of death 10.5 kilos.

Autopsy: No subcutaneous edema, no ascites; 3 c.c. urine showed much albumin and a few granular casts; kidneys about normal size, very pale, capsule slightly adherent, surface distinctly granular with fine and coarser nodules; cut surface pale and very opaque, cortex thin; liver and lungs congested, aorta apparently normal; heart large and flabby, many subendocardial hemorrhages; all chambers dilated.

Microscopical Examination: Moderate, diffuse, cellular proliferation of interstitial connective tissue with very marked periarterial fibrosis; moderate glomerular change; very extreme parenchymatous change with almost complete loss of nuclear staining; very marked desquamation, much exudate, and many hyaline casts.

Protocol 11.—Dog 11, 1910, a mongrel male, received two injections of 50 mg. and three injections of 30 mg. during forty weeks, and died eight days after the last injection; weight at beginning of experiment about 12 kilos, weight at time of death 9.9 kilos. Daily routine urine examinations after each of the first three injections showed the characteristic urinary findings of acute uranium poisoning, with much albumin and sugar, and many granular casts.

Autopsy: No subcutaneous edema or ascites; 10 c.c. urine showed much albumin and sugar, and many granular and cellular casts; kidneys rather small, pale, capsule slightly adherent, surface slightly granular, cut surface cloudy; cortex narrow; liver and lungs congested, aorta apparently normal; heart dilated in all chambers; weight 76 gm.

Microscopical Examination: Moderate diffuse proliferation of cellular interstitial connective tissue with little fibrosis excepting around the larger vessels. Moderate glomerular change; no glomerular cysts; very marked parenchymatous change, much desquamation in smaller tubules, much exudate and many casts.

Protocol 12.—Dog 12, 1910, a young terrier female, received one injection each of 30, 50 and 40 mg. of uranium nitrate during twelve weeks, and died four weeks after the last injection. Weight at beginning of experiment 10.5 kilos, maximum weight 15.8 kilos, weight at time of death 12.9 kilos.

Autopsy: No subcutaneous edema, ascites or urine; kidneys large, dark red, capsule slightly adherent, surface slightly granular, cut surface moist and cloudy, no cysts seen; liver and lungs congested, aorta apparently normal; heart dilated, numerous subendocardial ecchymoses in left ventricle; weight 85 gm.

Microscopical Examination: Moderate, diffuse and very cellular proliferation of interstitial connective tissue, with many large areas of round cell infiltration; relatively little glomerular change; few glomerular cysts; marked parenchymatous change, much flattening of tubular epithelium, some desquamation and much exudate; few casts seen.

Protocol 13.—Dog 15, 1910, a fox terrier female about 8 months old, received one injection each of 15 mg. and 20 mg. of uranium nitrate with an interval of nine weeks, and died eleven days after the last injection. Weight at beginning of experiment 5.3 kilos; weight at time of last injection 5.2 kilos, weight at time of death 3.6 kilos.

Autopsy: No subcutaneous edema and no ascites; 15 c.c. urine showed much albumin, moderate glycosuria, very many granular, and many waxy casts; kidneys swollen, congested, capsule not adherent, surface smooth, cut surface bulges, cortex wide and opaque; liver and lungs congested, aorta apparently normal; heart small, right ventricle dilated.

Microscopical Examination: Slight diffuse cellular proliferation of interstitial connective tissue; many areas of round cell infiltration; little glomerular change; some parenchymatous degeneration with little desquamation; no casts seen.

Protocol 14.—Dog 17, 1910, a fox terrier female about 8 months old, received two injections of 20 mg. of uranium nitrate with an interval of nine weeks, and died 11 days after the last injection. Weight at beginning of experiment 7.7 kilos, weight at time of death 5 kilos.

Autopsy: No subcutaneous edema and no ascites; 5 c.c. urine showed much albumin, some sugar, very many granular, and a few cellular casts; kidneys large, swollen, congested, capsule not adherent, surface smooth, cut surface bulges and is opaque; liver and lungs congested, aorta apparently normal; heart large, right side dilated, weight 50 gm.

Microscopical Examination: Slight, diffuse, cellular proliferation of interstitial connective tissue with rays extending almost to periphery in places; glomeruli show some exudate but little chronic change; very marked parenchymatous change, almost complete necrosis of epithelium, much desquamation and many casts.

Protocol 15.—Dog 19, 1910, a fox terrier female about 6 months old, received six injections of 10 mg. of uranium nitrate during eleven weeks, the first five being given during the first five weeks, and died five weeks after the last injection. Weight at beginning of experiment 3.7 kilos, weight at time of last injection 4.7 kilos, weight at time of death 2.5 kilos.

Autopsy: No subcutaneous edema and no ascites; 10 c.c. urine showed much albumin and sugar and very many granular and hyaline casts; kidneys small, pale, firm, capsule adherent, surface roughened, cut surface pale and opaque; liver, lungs and aorta apparently normal; heart small, not dilated, weight 26 gm.

Microscopical Examination: Slight cellular proliferation of interstitial connective tissue in inner zone of cortex with rays extending outwards to the surface; very little glomerular change; very marked parenchymatous change with almost complete necrosis of epithelium in many tubules; few casts.

Protocol 16.—Dog 22, 1910, a fox terrier female about 6 months old, received six injections of 5 mg. of uranium nitrate during eleven weeks, and died twenty-five weeks after the first injection; weight at beginning of experiment 4.4 kilos; weight at time of last injection 5.6 kilos; weight at time of death 4.3 kilos.

Autopsy: No subcutaneous edema and no ascites; 15 c.c. urine showed much albumin, some sugar and very many hyaline and partly granular casts; kidneys small, pale, capsule slightly adherent; surface smooth; cut surface cloudy and bulging; medulla darker than cortex; liver and lungs congested, aorta apparently normal; heart small, not dilated, weight 38 gm.

Microscopical Examination: Very marked, fairly diffuse cellular proliferation of interstitial connective tissue which in most places does not reach to the surface; moderate glomerular change, some exudate and some cysts; very marked parenchymatous change, marked fatty degeneration especially in Henle's loops; little desquamation and no casts.

Protocol 17.—Dog 24, 1910, a mongrel female about 6 months old, received seven injections of 5 mg. of uranium nitrate during thirty-one weeks, the first five being given during the first five weeks, and died nine days after the last injection. Weight at beginning of experiment 5.6 kilos, weight at time of last injection 6.9 kilos, weight at time of death 5.2 kilos.

Autopsy: No subcutaneous edema, ascites or urine; kidneys small, pale, capsule slightly adherent, cut surface cloudy and apparently more fibrous than normal; liver and lungs congested, aorta apparently normal; heart not dilated; weight 55 gm.

Microscopical Examination: Moderate, diffuse, cellular proliferation of interstitial connective tissue which does not quite reach the surface in most places; capsule moderately thickened; relatively little glomerular change, some exudate but no cysts; marked parenchymatous change, little desquamation, much exudate, and many dense, hyaline casts.

Protocol 18.—Dog 26, 1910, a young pointer, received two injections of 50 mg. of uranium nitrate with an interval of six and one-half weeks, and died seven weeks after the first injection; weight at beginning of experiment 20.5 kilos, weight at time of second injection 20.3 kilos, weight at time of death 12 kilos.

Autopsy: No subcutaneous edema or ascites. Five c.c. urine showed a trace of albumin but no casts; kidneys normal size, pale, capsule slightly adherent, surface smooth, cut surface cloudy, cortex appears rather reduced; liver, lungs and aorta normal; heart large, many subendocardial ecchymoses, no dilatation, weight 113 gm.

Microscopical Examination: Very marked cellular proliferation of interstitial connective tissue which is very diffusely distributed; kidney capsule markedly thickened; moderate glomerular change, much exudate and some small cysts; marked parenchymatous change, many tubules almost completely necrotic; moderate desquamation, much exudate and many dense hyaline casts. In places there is cystic dilatation of some of the tubules.

Protocol 19.—Dog 27, 1910, a young mongrel male, received two injections of 40 mg. of uranium nitrate with an interval of six and one-half weeks, and died twenty-four weeks after the first injection; weight at beginning of experiment 13.8 kilos, weight at time of second injection 16.7 kilos, weight at time of death 12 kilos.

Autopsy: No subcutaneous edema and no ascites; 15 c.c. urine showed much albumin and sugar and many granular casts; kidneys swollen and pale; capsule thick and firmly adherent, surface torn and finely granular with some coarser nodules, cut surface opaque, cortex thin with evident strands of connective tissue extending outwards to the surface; liver cloudy, lungs congested, aorta apparently normal; heart large, right ventricle dilated.

Microscopical Examination: Very extreme proliferation of interstitial connective tissue which involves practically the whole of the cortex, and is everywhere very cellular; very marked glomerular change, practically all glomeruli affected,

much exudate, proliferation and desquamation of capsular epithelium, and marked lobulation and shrinkage of tufts; a few glomeruli are hypertrophied; very extreme parenchymatous change in practically all tubules, with much dilatation of lumens, exudate, necrosis and desquamation; many tubules in areas of connective tissue proliferation are collapsed and atrophied; a few dense hyaline casts.

In discussing the conclusions which seem justifiable from the foregoing experiments, it will be more convenient to make a division under three headings: (1) edema formation; (2) renal changes; (3) cardiovascular changes.

EDEMA FORMATION

The use of uranium nitrate as an irritant with which to produce kidney lesions in animals was suggested by the reports of Richter,⁸ in which he described the production of edema in rabbits by the administration of uranium salts. In my first series of experiments on guinea-pigs¹ varying amounts of ascites and hydrothorax were found in some cases, but in no instance was there any subcutaneous edema or general anasarca. Pearce⁶ referred to this fact during a discussion of the experiments, and he suggested that more complete analogy to human nephritis might have been obtained if rabbits had been used for injection instead of guinea-pigs, since it had been shown that edema was so readily produced in rabbits during acute uranium intoxication, but the more recent experiments have shown that this is not the case.

In the series of sixteen experiments on rabbits which are here reported, but one animal showed any marked amount of fluid in the serous cavities, and only three others showed small amounts of ascites. In none of the cases was there any subcutaneous edema. It is true that three died of acute fibrino-purulent pleurisy, and that one showed a slight adhesive peritonitis, but these conditions were due to intercurrent infections and probably were in no way associated with any previous serous transudation.

The guinea-pig experiments showed vastly different results.⁹ "In the complete series of seventeen animals which survived for longer than three months, seven which died before the eighth month showed no edema, and of the ten which lived for longer than eight months, nine showed definite, and in some cases, marked edema. In four of the nine cases there was marked subcutaneous edema, which in three cases was associated with effusion into the serous cavities." All of the cases of subcutaneous edema occurred in animals to which relatively large doses of the drug had been administered; and, with one exception, in every case in which edema was present, the animal died within eleven days after the last injection.

8. Richter: Die experimentelle Erzeugung von Hydrops bei Nephritis. Beitr. z. klin. Med. Festschr. Herrn. Prof. Senator, Berlin, 1904, 283; and Experimentelle über der Nierenwassersucht, Berl. klin. Wehnschr., 1905, xlii, 384.

9. Dickson, E. C.: Edema Formation in Guinea-pigs in Chronic Experimental Uranium Nephritis. Proc. Soc. Exper. Biol. and Med., 1910, viii, 46.

The significance of these observations on guinea-pigs is at least two-fold. In the first place, it is definitely proved that edema formation in animals resulting from the administration of uranium nitrate is not confined to rabbits, and in the second place, a study of the conditions under which the edema was produced will show that there has been established a much closer analogy to the edema formation of Bright's disease in man.

In the production of experimental uranium edema in rabbits by Richter,⁸ Georgopoulos,¹⁰ Heinecke and Meyerstein¹¹ and others, the administration of uranium nitrate was always followed by forced water ingestion to produce an artificial hydremic plethora. In the experimental production of edema by combining the action of renal and vascular irritants, Pearce¹² also forced water ingestion, and he established as the three requisites for the production of renal edema, kidney damage, vascular damage and hydremic plethora. In my series of cases no attempt was made to induce edema by forcing water. Throughout the experiment the animals were kept in a large cage from which they were removed from time to time for weighing and for further injections. They were fed on a mixed diet in which was plenty of greens, and they obtained the whole of their water-supply from the green stuff which they consumed. A hydremic plethora due to increased water ingestion is obviously impossible under such conditions, and one can only assume that the increase in the fluids in the body must have been due to the suppression of urine which occurred during the terminal attack of acute nephritis.

The conclusion of all observers who have employed uranium salts for the experimental production of edema is that there must be some specific action of uranium in addition to its effect on the renal epithelium, which is at least partly responsible for the edema formation. The observations of Heinecke and Meyerstein,¹¹ who found that edema was produced in rabbits which were poisoned with chromates only after the serum of rabbits suffering from acute uranium poisoning had been injected intravenously, were very suggestive, even though the results were not constantly produced, and there seemed to be sufficient ground for their conclusion that there must be some vascular damage, in addition to the renal damage, which was responsible for the edema formation. The oncometer experiments of Schlayer and Hedinger¹³ and Takayasu¹⁴ gave

10. Georgopoulos: Experimentelle Beiträge zur Frage des Nierenwassersucht. *Ztschr. f. klin. Med.*, 1906, lx, 411.

11. Heinecke and Myerstein: Experimentelle Untersuchungen über die Hydrops bei Nierenkrankheiten. *Deutsch. Arch. f. klin. Med.*, 1907, xc, 101.

12. Pearce: The Production of Edema; an Experimental Study of the Relative Etiologic Importance of Renal Injury, Vascular Injury and Plethoric Hydremia. *THE ARCHIVES INT. MED.*, 1909, iii, 422.

13. Schlayer and Hedinger: Experimentelle Studien über toxische Nephritis. *Deutsch. Arch. f. klin. Med.*, 1907, xc, 1.

14. Schlayer, Hedinger and Takayasu: Ueber nephritisches Oedem. *Deutsch. Arch. f. klin. Med.*, 1907, xci, 59.

the first experimental proof that such must indeed be the case, and their observations have been confirmed by Pearce and his assistants.¹⁵ Pearce¹² has further established the importance of vascular injury in renal edema in the series of experiments to which I have already referred, namely, those in which he employed combinations of various vascular irritants with bichromate poisoning and forced water ingestion.

In view of these uniform conclusions as to the importance of damage to the blood-vessels in the production of renal edema, but one explanation can be given for its occurrence in the cases here described. The fact that the death of the animals and the onset of the edema occurred during the acute intoxication which followed the last injection of uranium, indicates definitely that the edema was a true renal edema, and not edema due to failing compensation of an overworked heart; and the appearance of the hearts at autopsy confirmed this view. There must therefore have been some condition present after the several months of treatment, which rendered the animals unable to cope with the hydremia of an acute attack of nephritis which was presumably no more severe than that of any one of the several previous attacks. It is difficult to see how this condition could be other than some damage to the smaller blood-vessels, especially in those cases in which the subcutaneous edema was so well developed. I believe we are justified in concluding that the repeated injections of uranium had so acted on the (blood) vascular system that it was no longer able to withstand the increase in the volume of blood which results from the partial or complete suppression of urine of acute uranium poisoning.

Whether or not this be the correct explanation, there can be no doubt that the formation of edema, occurring as it did in these animals during an attack of nephritis, and without any artificially produced hydremic plethora, establishes another step in the advance toward a complete analogy to Bright's disease in man.

RENAL CHANGES

The histological appearance of chronic uranium nephritis has already been described in the beginning of this report, and the characteristic type of the lesions, as well as the constancy with which they occur, has been mentioned. In order to determine whether similar lesions of spontaneous origin occur in normal animals, a series of routine histological examinations was made of the kidneys of fifty rabbits and 100 guinea-pigs which had been sacrificed for various purposes in the laboratory. The results of these observations have recently been reported by Ophüls,¹⁶ from whom I shall quote the following:

15. Pearce, Hill and Eisenbrey: *Experimental Acute Nephritis; the Vascular Relations and the Elimination of Nitrogen*. Jour. Exper. Med., 1910, xii, No. 2, 196.

16. Ophüls, W.: *Occurrence of Spontaneous Lesions in Kidneys and Livers of Rabbits and Guinea-Pigs*. Proc. Soc. Exper. Biol. and Med., 1911, viii, 75.

In view of the importance of the occurrence of spontaneous lesions in the kidneys and in the livers of rabbits and of guinea-pigs in reference to the experimental work on these organs, Dr. E. C. Dickson and myself have made a careful study of these organs and incidentally of the heart and of the aorta in fifty rabbits and in one hundred guinea-pigs. The animals used were partly fresh animals from the market, partly animals raised at the laboratory. * * *

Twenty-eight rabbits, among them some of the old ones, had entirely normal kidneys, nine showed slight parenchymatous lesions, three a few small areas of cellular infiltration. In ten we found scattered small areas in which were marked interstitial lesions with the formation of small depressions on the surface. Four of these proved to be radially arranged chronic septic foci which extended from the vicinity of the papilla to the outer surface. In the other six cases the same arrangement of the newly-formed connective tissue in narrower radial bands which start deep in the pyramids was noticeable although no evidence of a septic infection could be discovered. The interstitial lesions were accompanied by marked epithelial lesions in two cases only, producing to a certain extent the picture of a chronic parenchymatous nephritis, although the lesions were never very extensive. Lesions of the blood-vessels or primary lesions of the glomeruli were not found in any case. * * *

Of the one hundred guinea-pigs the large majority had been injected with sediment from urine or other material suspected of containing tubercle bacilli; only a few actually developed tuberculosis. There were several especially selected old animals. Sixty-three of the one hundred had absolutely normal kidneys, two showed slight parenchymatous lesions and in thirty-five there were scattered small areas of cellular infiltration in the cortex which were sometimes rather numerous; at other times they were found with difficulty only on careful examination of several sections. They consist largely of lymphocytic cells and a few larger cells with vesicular nuclei. They do not seem to have much tendency to develop into fibrous tissue although occasionally such a change was observed but always to a limited degree only. * * * In few cases fibrous thickening of the capsules of glomeruli and cystic dilatation of the same were observed as spontaneous lesions similar to certain types of glomerular cysts described by Dickson in his paper on the "Experimental Production of Chronic Nephritis in Animals By the Use of Uranium Nitrate." These were, however, rare exceptions, whereas in uranium poisoning such glomerular lesions seem to be nearly constant. Large scars or lesions of the blood-vessels were not found in any case.

It will be seen from this extract that, although there was a fairly high percentage of cases which showed spontaneous kidney lesions, the type of the lesions which were found was entirely different from that of chronic uranium nephritis. Indeed, in some of the rabbits of my series there was quite evidently a combination of the two conditions, for in kidneys which were distinctly of the uranium type a few scattered foci could be recognized as more closely resembling the spontaneous type of lesions. And, moreover, although the spontaneous lesions which were found in the kidneys of rabbits were entirely different from those which were found in guinea-pigs, the lesions which were produced by uranium in both rabbits and guinea-pigs, and indeed also in dogs, were so similar as to be almost identical in many cases.

In the dog experiments, too, care was taken to exclude as much as possible the occurrence of spontaneous chronic kidney lesions. Young, healthy animals were always selected for experimentation, and of the nineteen cases here reported, fifteen were known to be less than 9 months

of age at the time of the first injection. In the 1909 series, Protocols 1 to 5,¹⁷ especial precautions were taken to preclude the possibility of the development of spontaneous lesions. Puppies were weaned in the laboratory and were placed on a diet consisting of mush, bread and milk, with an occasional feeding of dried, chopped meat from which the extractives had been removed in the soup-pot. The animals thrived on this diet and received no other food during the whole course of the experiment. When they were 5 months old the series of injections of uranium nitrate were commenced, and, with the exception of one animal which was killed in a fight fourteen weeks after injection, they were allowed to live for from twenty-seven to thirty-four weeks after the first injection. The oldest of these puppies was 11½ months old when killed, so that in every case the experiment was terminated within the first year of life of the animal. The carefully regulated diet was such that the kidneys were protected as much as possible from the influence of any irritating substance excepting the uranium nitrate, and, when we consider in addition to this that in every case the animal was less than 1 year old at the end of the experiment, there seems little doubt that the lesions which were found must have been due to the administration of the uranium nitrate. Further experiments have confirmed this conclusion, for in the whole series of dogs which were observed during the following eighteen months, not one animal failed to show kidney lesions of a chronic nature, and of the uranium type, excepting those which died during the initial acute intoxication. The experiments show definitely that dogs are well adapted for the production of chronic kidney lesions, and that these lesions may be produced in practically all cases by the administration of uranium nitrate.

Although the occurrence of the chronic lesions in the kidneys has been so constant, the method by which these lesions are produced has not been revealed. A very striking feature of chronic uranium nephritis is that, even in the advanced cases in which the kidneys are hard and granular, and in which the greater part of the parenchyma is replaced by dense fibrous tissue, there is no demonstrable lesion in the vessels of the kidneys or of the peripheral vascular system. It is true that in many cases, especially in dogs, there is a peculiar peri-arterial development of fibrosis, and that in rabbits many of the glomerular tufts show almost complete fibrosis, but in the capillaries of these tufts the only evidence of lesion is a slight thickening of the walls, and the deposit of some hyaline substance around them, and in the larger arteries no lesions whatever can be demonstrated either in the intima or in the media. This fact is especially interesting in view of the recent experiments of Harvey¹⁸ who, by the

17. The 1909 series of experiments on dogs was reported before the California Academy of Medicine in December, 1909. *Cal. State Jour. Med.*, 1910, viii, No. 4, 140.

18. Harvey: Auto-intoxication and Experimental Nephritis in Rabbits. *Jour. Path. and Bact.*, 1911, xvi, No. 1, 95.

administration of para-hydroxy-phenyl-ethylamine, was able to produce chronic lesions in the kidneys of rabbits secondarily to the development of atheroma of the aorta and arteriosclerosis of the renal artery and its branches. The kidneys which Harvey describes are undoubtedly of the arteriosclerotic type, and his experiments would seem to prove that the etiology of that type of nephritis is entirely different from that of such types of atrophied and granular kidneys as are represented by uranium chronic nephritis.

Another point of interest in these cases is one to which I referred in my previous report. In the kidney lesions of acute uranium poisoning the tubular epithelium is the part which shows the most marked anatomical change. With the exception of the formation of the hyaline bodies which Christian³ has described in the glomerular tufts during acute uranium nephritis, no characteristic lesions have been found in the glomeruli, yet in the kidneys of chronic uranium nephritis the glomerular lesions are varied and severe. The most common, as well as the most extensive changes, are seen in the glomerular capsule, with its cystic dilatation, proliferated epithelium and hyaline basement membrane. The tufts also are more or less extensively damaged. In the less severe cases we find fragmentation of the nuclei and often some evidence of increase in connective tissue cells; and in the advanced cases there is distinct fibrosis and lobulation of the tufts, and, in rabbits, the thickening of the walls of the capillaries and the deposit of some hyaline substance in a part or the whole of the tufts, to which reference has already been made. Moreover, the compensatory hypertrophy of the more normal appearing glomeruli indicates diminution in function of the damaged glomeruli.

Still another interesting feature in chronic uranium nephritis is the characteristic distribution of the newly formed connective tissue. In every case it is more marked and much more fibrous in the inner zone of the cortex, and in the early cases it is in this region, especially around the larger blood-vessels, that the proliferation of connective tissue is first seen. Reference has already been made to the peculiar peri-arterial fibrosis which occurs around the larger arteries. From this deeper, more fibrous zone, the connective tissue extends outward toward the surface of the kidney, usually following the lines of the medullary rays. In those cases in which the surface of the kidney is granular, the processes extend to the surface and correspond to the small depressions on it, but in many cases where the surface is still quite smooth histological examination shows in the deeper parts much newly formed connective tissue from which many radiating processes extend into the outer zone of the cortex but do not reach the surface. In some kidneys there is also a diffuse connective tissue proliferation which more or less completely destroys the kidney cortex between the rays; but this seems to be a later stage in the process, and is found only in the most severe cases. Occasionally there is also a

more or less marked extension of the connective tissue proliferation into the medulla, but this is not at all constant.

A very striking feature of the connective tissue proliferation is that all the different stages are represented in the kidneys of animals which have not received any injections for several months. The condition is not that of a quiescent scar formation, for, although in the deeper portions the newly formed connective tissue is densely fibrous and contains but few nuclei, in the peripheral portions it is usually very cellular, with many spindle shaped cells with large, clear vesicular nuclei, and with many round plasma cells, clearly indicating that the connective tissue proliferation is still progressive. Only in those cases in which the kidney is markedly shrunken and granular does the peripheral portion of the connective tissue have a dense fibrous structure, and even in these there are usually scattered areas of cellular infiltration in which the cells consist partly of fibroblasts and partly of plasma cells.

The explanation of these phenomena is not clear. At first sight it might seem that the extreme degeneration and necrosis of the tubular epithelium in acute uranium poisoning must be responsible for the secondary proliferation of the connective tissue and for the marked glomerular changes. If we accept the teaching of Senator, namely, that destruction of any part of a tubule system will lead to secondary destruction of the remainder of that tubule system, we could explain the destruction of the glomeruli, and, perhaps also, the cystic dilatation of the glomerular capsules. But it is difficult to understand how simple destruction of the tubular epithelium could account for the peculiarly constant development of connective tissue in the inner zone of the cortex, or for the evident progression of the connective tissue proliferation which is found months after the insult which produced the primary necrosis. Again, if it is simply the epithelial necrosis which is responsible, one would expect to find equally severe secondary fibrosis in kidneys which have been poisoned with potassium bichromate, for in acute bichromate nephritis the lesions are practically indistinguishable from those which are produced by uranium nitrate. The experiments of Ophüls seem to show that this is not the case. He subjected forty guinea-pigs and fifty rabbits to repeated injections of potassium bichromate during periods varying from a few weeks to almost two years' duration. In his recent report¹⁹ on the effects produced in guinea-pigs he says:

The conclusion seems justified that it is impossible to produce severe lasting lesions in guinea-pigs with chromates, probably because in these animals the chromates are too toxic in a general way, and too slightly effective on the kidneys locally.

19. Ophüls, W.: Experimental Nephritis in Guinea-Pigs by Subcutaneous Injections of Chromates, *Proc. Soc. Exper. Biol. and Med.*, 1911, ix, 13.

And in describing his experiments on rabbits he says:²⁰

Rabbits are relatively more susceptible to the action of chromates than guinea-pigs. * * * The extreme lesions which may develop after a while as a result of crowding of large doses were described by me in 1908 (*Jour. Med. Research*, 1908, xviii, 49), but they do not necessarily follow even repeated administration of large doses. * * * Fifty animals were experimented upon. The dosage varied between two centigrams and two milligrams. Some of the animals were kept alive for a year or more. So far as epithelial lesions are concerned the experiences are similar to those in guinea-pigs, except that the epithelial lesions became more severe and seemed to continue longer after the last injection. Very marked interstitial lesions were observed at times, but they resemble those observed spontaneously so closely, and occurred so irregularly, sometimes soon, sometimes late after the administration of various doses and sometimes not at all, that any definite conclusion of their relation to the injections could not be arrived at.

It is true that the findings of Smith² differ considerably from those described by Ophüls. From a series of thirty-nine rabbits and seven guinea-pigs which survived for longer than three months after the initial injection of either potassium bichromate or uranium nitrate (the exact number of each is not stated), he concludes that "the chronic lesions caused by uranium nitrate and potassium bichromate were the same," and that "bichromate of potassium produces more fibrous tissue formation with macroscopic depressions on the surface of the kidney than uranium nitrate."

If these observations of Smith are based on a sufficiently large number of cases of bichromate nephritis, his results are of great interest in view of the large number of negative results described by Ophüls, and they would greatly weaken the argument that in uranium nephritis it is not alone the epithelial necrosis which is responsible for the secondary development of fibrous tissue. However, judging from the large number of experiments which have been conducted in our laboratory, there seems little doubt that in uranium nitrate nephritis there is some factor which is lacking in potassium bichromate nephritis, and which is responsible for the almost constant production of chronic kidney lesions in the former. Just what this factor may be is still in doubt. I am convinced that there is something more than mere coincidence in the fact that there is the occurrence of edema in acute uranium intoxication, and the development of chronic kidney lesions after prolonged uranium administration. If it be true that the formation of edema depends on a combination of lesions in the kidneys and in the peripheral blood-vessels, I believe there is no doubt that the development of the chronic kidney lesions is dependent on a similar combination of lesions. Indeed, I believe that no other condition than some damage to the vascular system can explain

20. Ophüls, W.: Experimental Nephritis in Rabbits by Subcutaneous Injections of Chromates. *Proc. Soc. Exper. Biol. and Med.*, 1911, ix, 13.

the peculiarly characteristic distribution and the progressive type of interstitial connective tissue proliferation which we find in chronic uranium nephritis.

THE CARDIOVASCULAR CHANGES

The question of the interrelationship which exists between hypertrophy of the left ventricle of the heart and chronic interstitial nephritis is one which has received much attention. Various observers have approached the problem by simulating the local kidney conditions of chronic interstitial nephritis by the actual reduction of kidney substance by surgical methods. Bradford,²¹ Passler and Heinecke,²² Janeway,²³ Sampson and Pearce²⁴ and others have found that, following such reduction of kidney substance, there develops in certain cases a rise in blood-pressure and hypertrophy of the left ventricle of the heart. Rautenberg²⁵ also by mechanically damaging one kidney and subsequently removing the other kidney has succeeded in raising blood-pressure in rabbits to such an extent that aneurysms of the aorta are formed. But the number of observations of development of hypertrophy of the heart in cases of experimental chronic nephritis is very small. Charcot and Gombault²⁶ and Levaditi²⁷ report hypertrophy of the left ventricle of the heart in cases of experimental chronic nephritis produced by lead salts and vinylamin, respectively. Siegel²⁸ reports hypertrophy of the left ventricle of the heart in three dogs which had been subjected to uranium injections for from three to six weeks, and Smith² reports four cases of apparent heart hypertrophy in his series of uranium nitrate rabbits. However, the limited number of instances in which experimental chronic nephritis has been produced has of necessity limited the number of observations which could be made.

In a series of experiments such as those reported in this paper the difficulty of determining the presence of a true heart hypertrophy is very

21. Bradford: The Results following Partial Nephrectomy, and the Influence of the Kidney on Metabolism. *Jour. Physiol.*, 1899, xxiii, 415.

22. Passler and Heinecke: Versuche zur Pathologie des Morbus Brightii. *Verhandl. d. deutsch. pathol. Gesellsch.*, 1905, ix, 99.

23. Janeway: Note on Blood-Pressure Changes Following Reduction of the Renal Arterial Circulation. *Proc. Soc. Exper. Biol. and Med.*, 1909, vi, 109.

24. Sampson and Pearce: A Study of Experimental Reduction of Kidney Tissue with Special Reference to the Changes in That Remaining. *Jour. Exper. Med.*, 1908, x, 745.

25. Rautenberg: Erzeugung chronischer Nierenkrankungen mit folgender Blutdrucksteigerung und Arteriosklerose. *Deutsch. med. Wchnschr.*, 1910, xxxvi, 551.

26. Charcot and Gombault: Note relative à l'étude anatomique de la néphrite saturnine expérimentale. *Arch. de physiol.*, 1881, Series 2, viii, 126.

27. Levaditi: Experimentelle Untersuchungen über die Necrose der Nierenpapillæ. *Arch. internat. de pharmacodyn.*, 1901, viii, 5.

28. Siegel: Über experimentelle Nephritis. *Verhandl. d. Congresses Inn. Med.*, 1907, xxiv, 217.

great. In normal animals quite definite ratios of heart to body weight have been established, and in animals which have been subjected to experimentation but in which the weight at the time of death shows no marked variation from the weight during the time in which the animal has been under observation, similar heart to body ratios can be established and compared with those of normal animals. But in those animals which lose in weight during the course of the experiment, and especially in animals which decrease rapidly in weight during the terminal illness, such heart to body ratios are difficult to determine. One cannot tell exactly what proportional loss of weight occurs in the heart itself during the progressive loss in the whole body weight; in such cases one must rely largely on the macroscopic appearance of the heart. In well-marked cases of heart hypertrophy there is little difficulty in forming an opinion, but in doubtful cases, and especially in the smaller animals, one must be very guarded in making the decision that hypertrophy actually exists.

Conservative inspection of the hearts of the animals reported in this paper showed quite definitely that there was hypertrophy of the heart, especially of the left ventricle, in two guinea-pigs, four rabbits and nine dogs. In several other cases the heart gave one the impression of being hypertrophied, but the hypertrophy, if present, was too slight to justify a definite conclusion. In the rabbits and guinea-pigs no confirmatory heart to body ratios were established, but in the dogs such ratios were determined and were studied in comparison with those worked out in normal dogs by Stewart²⁹ and Joseph.³⁰ Table A shows clearly the result of this comparison.

In Stewart's series of twenty normal dogs he found that there was an average heart to body ratio of 0.00724, a ratio approximately the same as that obtained by Joseph, namely, 0.00750. But among Stewart's twenty animals there were three which showed a ratio of 0.00800 or over, and in his series of experiments he withheld a diagnosis of heart hypertrophy in all cases in which the ratio did not exceed the maximum ratio obtained in his normal dogs. It will be seen that some of my cases fall below the maximum normal ratio as estimated by Stewart, and may therefore be viewed with some suspicion. But when we consider that in all my cases the animals were confined to small quarters during practically the whole of their lifetime, and would presumably on that account show relatively smaller hearts than would active street dogs, the constantly high ratio is very striking. And it is of interest that my three normal controls, which were kept under similar conditions, all showed heart to body ratios which were well below the average as established by Stewart. It is also

29. Stewart: An Experimental Contribution to the Study of Heart Hypertrophy. *Jour. Exper. Med.*, 1911, xiii, 187.

30. Joseph: The Ratio between the Heart Weight and the Body Weight in Animals. *Jour. Exper. Med.*, 1908, x, 521.

of interest that in all but two of the cases included in the accompanying table, the weight at the time of death was as great or greater than at any time during the course of the experiment, due to the fact that these animals were intentionally or accidentally killed several weeks after the last injection. In two cases in which there was definite loss of body weight immediately preceding death, the ratio of the heart weight at the time of death to the maximum body weight during life was high, and I believe that one is justified in assuming that the actual heart to body ratio must lie somewhere above that ratio, but below the ratio of the heart weight at time of death to the body weight at time of death. Accordingly in the table these two ratios are entered as (a) and (b) and the actual heart to body ratio is assumed to lie somewhere between the two, in both cases at a point which is high as compared with the average normal ratio.

TABLE A.—(A) SHOWING THE RATIOS WHICH EXIST BETWEEN THE HEART WEIGHT AND THE BODY WEIGHT IN DOGS WITH CHRONIC URANIUM NEPHRITIS;
(B) SHOWING SIMILAR RATIOS IN NORMAL DOGS

Protocol Number	Animal Number	Duration of Exp't, Weeks	Body Weight in Grams	Heart Weight in Grams	Ratio
2	3—1909	34	10,260	80	0.00779
3	4—1909	31	14,280	118	0.00802
5	6—1909	29	5,610	60	0.01069
6	1—1910	61	7,680	61.5	0.00800
7	2—1910	71	16,300	145	0.00888
8	3—1910	60	21,600	180	0.00818
9	5—1910	25	(a) 8,980	73	0.00822
..		..	(b) 8,020	73	0.00922
17	24—1910	32	(a) 6,900	55	0.00790
..		..	(b) 5,200	55	0.01041
NORMAL CONTROL ANIMALS					
..	8—1909	..	8,640	60	0.00694
..	4—1910	..	8,400	60	0.00714
..	29—1910	..	4,680	34	0.00705
Stewart's Series—20 dogs					0.00721
Joseph's Series					0.00750

These weights and ratios are presented with the full realization that in some of them the proportional weight of the heart is somewhat below what may be considered the limit of normal variation. However, I believe that when we take into consideration the age of the animals which were used, the conditions under which they were kept, and the fact that in every case the heart showed macroscopic appearance of hypertrophy, one is justified in presenting them as evidence of proportional increase in the size of the heart.

The condition of the blood-vessels has already been described in considering the lesions which are found in the kidneys.

CONCLUSIONS

1. By the administration of uranium nitrate it is possible to produce a chronic diffuse nephritis in guinea-pigs, rabbits and dogs.

2. The histological picture of the lesions is characteristic, and is analogous to, but not identical with, that found in chronic diffuse nephritis in man.

3. The chronic lesions, even in the most severe cases, are not associated with demonstrable arterial lesions, although the mode of development suggests that there must be some functional vascular damage.

4. After prolonged administration in guinea-pigs a terminal attack of acute nephritis is frequently accompanied by more or less marked anasarca.

5. Associated with the more severe kidney lesions there is frequently a more or less marked hypertrophy of the left ventricle of the heart.

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