UNIVERSITY OF TORONTO STUDIES

PATHOLOGICAL SERIES

No. 4: REPORT OF THE EXPERIMENTAL PRODUCTION OF CHRONIC NEPHRITIS IN ANIMALS BY THE USE OF URANIUM NITRATE, BY ERNEST C. DICKSON

(REPRINTED FROM ARCHIVES OF INTERNAL MEDICINE, JUNE, 1909.

THE UNIVERSITY LIBRARY: PUBLISHED BY THE LIBRARIAN, 1909

University of Toronto Studies

COMMITTEE OF MANAGEMENT

Chairman: Robert Alexander Falconer, M.A., Litt.D., LL.D., D.D.
President of the University

Professor W. J. Alexander, Ph.D.

Professor W. H. Ellis, M.A., M.B.

Professor A. Kirschmann, Ph.D.

Professor J. J. Mackenzie, B.A.

Professor R. Ramsay Wright, M.A., B.Sc., LL.D.

Professor George M. Wrong, M.A.

General Editor: H. H. Langton, M.A.
Librarian of the University

A REPORT OF THE EXPERIMENTAL PRODUCTION OF CHRONIC NEPHRITIS IN ANIMALS BY THE USE OF URANIUM NITRATE.*

ERNEST C. DICKSON, B.A., M.B.
SAN FRANCISCO.

INTRODUCTION.

The study of chronic nephritis is one which for many years has commanded the interest and investigation of many observers. Since 1827, when Richard Bright first described the condition which we now know as Bright's disease, many attempts have been made to determine the relationship existing between the various types of diseases of the kidney, and the conditions which influence the development of those different types. It is impossible to discuss here the immense amount of literature which has been written on this subject, or to consider the bitter controversies which have waged. Even to-day, although certain types of subacute and chronic conditions are universally recognized, there is great difference of opinion as to the etiologic factors at work in the development of each.

In 1879 Weigert[1] published an elaborate discussion of kidney diseases viewed from the standpoint of pathologic anatomy, and in his classification he described a series of kidneys as "chronic hemorrhagic with heart hypertrophy" in which there are definite interstitial changes associated with the parenchymatous degenerations. This group he subdivides into three smaller groups:

1. Those kidneys which are normal or slightly larger than normal in size, are red or mottled in color and are firmer in consistency than normal. The capsule strips easily, the cut surface bulges a little on section, and the cortex is pale or mottled in color, while the pyramids are darker. There is a certain amount of fatty degeneration of the epithelium of the convoluted tubules, many of which are more or less completely obliterated while others are dilated and contain a clear or

* From the Pathological Laboratory of the University of Toronto, Canada.

* A preliminary report of these experiments was read before the Association of American Pathologists in Washington, D. C., in June, 1907, and before the Laboratory Section of the Canadian Medical Association in Montreal, P. Q., in August, 1907.

1. Weigert: Die Bright'sche Nierenkrankungen von pathologisch-anatomischen Standpunkte. Samml. klin. Vortr. (Volkmann's), 1879, clxii-clxiii, 1411.

granular exudate in their lumens. The collecting tubules and the loops of Henle contain casts. The glomeruli may be unchanged or may show thickening of the capsules, and there may be connective tissue change in the tufts. There is some proliferation of the intertubular connective tissue, and there may or may not be thickening of the intima of the arteries.

2. Forms which are similar to those just described, but which show in addition definite macroscopic granulations on the surface. There is greater glomerular and tubular change and much more new growth of connective tissue. Clinically this type contrasts with the chronic interstitial, in its shorter duration, the greater amount of albumen, and the presence of edema. It may be considered as a further stage in the process of which the type last described forms a part.

3. Large white kidneys which closely resemble the above type both clinically and histologically except that they are anemic and show much more fatty change.

Weigert also refers to kidneys which are atrophied and granular, with very marked degeneration of the glomeruli and tubules, with the formation of cysts and with calcium deposit, and with marked diffuse increase of the connective tissue. This group he divides into two smaller groups: (a) the small red atrophic kidney; (b) the small white atrophic kidney. The condition is characterized clinically by the long duration, the absence of edema, and the hypertrophy of the left ventricle of the heart.

In discussing the etiology of chronic nephritis he points out that a parenchymatous nephritis, i. e., one in which the parenchymatous change is the most marked but which shows also some interstitial change, can pass over into the stage of a shrunken kidney. He also believes that all new formation of connective tissue in the kidney is invariably preceded by degeneration of the tubules or glomeruli.

In 1897 Senator[2] made a definite division of contracted kidneys into two groups, primary and secondary. The primary sclerotic, together with the arteriosclerotic, form by far the greater number of cases of chronic nephritis, and are progressive from the beginning without any primary acute inflammation, and often without acute exacerbations. The secondary contracted kidney on the other hand, dates from some acute infection, and has an initial acute stage which may be considered as a true parenchymatous nephritis. He believes

2. Senator: Die Pathogenese der chronischen Nephritis. Berl. klin. Wchnschr., 1897, xxxiv, 820.

that this type is also progressive because of some disturbance of the vascular system.

Samuel West[3] in his Lettsonian Lectures in 1899 admits that viewed from a pathologic standpoint there is a possibility of a progression from an acute parenchymatous inflammation through the stages of the subacute to the small granular or atrophic kidney; but he believes that such a process is practically never found clinically. He believes it as reasonable to suppose that the so-called primary parenchymatous nephritis is an acute exacerbation of a pre-existing chronic

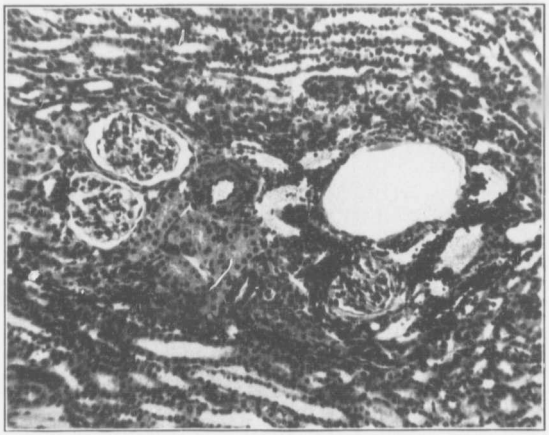

Fig. 1.—Guinea-pig 25: 46 injections of 0.25 mg. in 77 days. Dilatation of tubules in the medullary rays, cellular infiltration between tubules, desquamated cells in the dilated lumens, and glomerulus with thickened capsule surrounded by round-cell infiltration.

condition, as that it is the initial lesion upon which the subsequent granular condition depends.

Freidrich Müller[4] in 1904 points out that acute parenchymatous nephritis due to many infections may eventually go on to the develop-

3. West: Some clinical aspects of granular kidney. Brit. Med. Jour., 1899, i, 329.

4. Müller: Morbus Brightii (Korreferat). Verhandl. d. deutsch. path. Gesellsch., 1905, ix, 64.

ment of a contracted or indurated kidney. He believes that many of the so-called idiopathic or primary indurated kidneys are thus classified because a sufficiently careful search for a definite causative factor has not been made. He believes that while in many cases of nephritis following acute infection there may be a long-drawn-out and progressive condition with slight disturbances of temperature and with periodic exacerbations, yet in a certain number of such cases there is a complete cessation of all symptoms and freedom from any disturbance for many years. In such cases he considers that the condition is that of healing by scar formation, and that it is not progressive, although a kidney so damaged must necessarily offer a focus of lowered resistance to subsequent insult. He compares the lesion to that found in the endocardium after a healed valvular endocarditis, or to that condition of fibrosis which is found in the lung after an unresolved pneumonia. Although the clinical course of this type differs from that of the so-called idiopathic chronic nephritis in the acute onset with the presence of albumen and casts, and in the gradual clearing up of the acute symptoms with the subsequent development of polyuria and hypertrophy of the left ventricle, yet histologically the kidneys can not be distinguished from one another.

In 1906 Lohlein[5] showed that in a certain number of patients who succumbed to chronic nephritis there was a definite history of a preexisting infection followed by a quiescent period of several years before the appearance of the terminal symptoms. He based his conclusions on a study of scarlatina and diphtheria in which he found that glomerular change played a very important part in the nephritis following these diseases. He says that advanced fibrosis with hyaline change of the tufts and thickening of the capsules of the glomeruli may undoubtedly date back to a glomerular nephritis in a certain number of young subjects in which the history definitely locates the primary attack and excludes probability of subsequent irritation to the kidneys.

In view of such differences of opinion resulting from purely clinical observations, an attempt has been made to produce a chronic nephritis in animals by experimental means, in the hope that some condition might be produced analogous to that found in man, and that from the method of experimental production some light might be thrown on the etiologic process which produces the condition in

5. Lohlein: Ueber die entzündlichen Veränderungen der Glomeruli der menschlichen Nieren und ihre Bedeutung für die Nephritis. Verhandl. d. deutsch. path. Gesellsch., 1906, 217.

man. Uranium nitrate was the irritant selected, because Richter[6] has recently shown that by the use of this drug "it has become possible to produce a diseased condition in animals which is analogous throughout to Bright's disease." The experiments were performed in the Pathological Laboratory of the University of Toronto, under the direction of Professor J. J. MacKenzie. I wish here to express my gratitude to Professor MacKenzie for the interest with which he has followed the work, and for the many helpful suggestions he was at all times ready to give. I am also indebted to Dr. V. E. Henderson for help and advice throughout the course of the experiments.

HISTORICAL.

A review of the literature on experimentally produced chronic nephritis forces one to the conclusion that the results obtained have not been very satisfactory. In 1904 Rose Bradford[7] suggested that a possible explanation of the many failures might lie in the fact that up to that time no irritant was known by which one could produce in animals a condition analogous to acute Bright's disease in man, i. e., an acute parenchymatous nephritis associated with the formation of edema. An exhaustive review of the literature up to January, 1907, may be found in Lyon's report[8] entitled, "An Experimental Study on the Action of some Poisons and Toxins on the Kidney and Spleen," and in Ophüls' article[9] on "Experimental Chronic Nephritis." Space will not permit more than a very brief review at this time.

Bradford[7] has stated that mere destruction of renal parenchyma by a metallic poison is not sufficient to lead to the production of a true contracted kidney. Potassium bichromate injected directly into the kidney of an animal through the renal vein causes an extensive destruction of the renal epithelium, and if the animal survives the first acute attack, a subsequent atrophy of the tubules and glomeruli with definite shrinking of the kidney. But the condition has not the picture of a true contracted kidney because there is no proliferation of connective tissue.

---

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

7. Bradford: On Bright's disease and its varieties. Croonian Lectures, 1904. Lancet, London, 1904, clxvii, 191.

8. Lyon: Experimental study on the action of some poisons and toxins on the kidney. Jour. Path. and Bacteriol., 1903-04, ix, 400.

9. Ophüls (W.): Experimental chronic nephritis. Jour. Am. Med. Assn., 1907, xlviii, 483.

Lyon[8] conducted an extensive series of experiments in which he produced acute nephritis in animals by the use of different poisons and toxins. But in so far as the production of anything like a chronic condition was concerned his results were not very satisfactory. He succeeded in obtaining some new formation of connective tissue around some of the large deposits of calcium which occurred in the medulla of kidneys poisoned with corrosive sublimate, but he was unable to find any atrophied glomeruli with fibrous thickening of the capsule even in rabbits that had undergone a chronic poisoning which extended over a period of one hundred and ten days. In his summary he says: "In no case and by no variation of experimental method

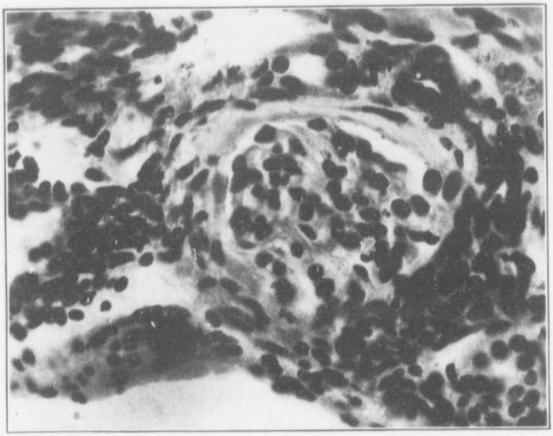

Fig. 2.—Guinea-pig 25: High power of glomerulus showing thickening of capsule and infiltration of round cells around it.

have I been able to produce and follow the evolution of changes at all analogous to those which we find in sub-acute and chronic diffuse nephritis in man."

Petroff[10] in his experiments on the effects of various metals on the kidneys, found that when the action of the irritant was continued for a considerable length of time, there resulted necrosis of the renal

10. Petroff: Ueber die Einwirkung der Metalle auf die Nieren. Diss., Würzburg, 1905.

epithelium with definite proliferation of connective tissue. But at no stage of the experiments did he find albumin or casts in the urine.

Ehrlich[11] and Levaditi[12] were somewhat more successful, for by the use of vinyl amine they were able to produce a definite fibrosis of the kidneys in white mice, which was secondary to localized hemorrhagic necrosis of the papillæ. This condition in one case was associated with characteristic urinary findings, as well as with hypertrophy of the left ventricle of the heart and albuminuric retinitis.

Pässler and Heineke[13] have recently reported a series of experiments by which they simulated the local conditions of chronic nephritis by directly destroying the greater part of the parenchyma of one kidney, and subsequently excising the other kidney. In certain animals which survived there resulted polyuria and hypertrophy of the left ventricle, and associated with this was the peculiar cachectic condition of chronic nephritis.

Ophüls[9] reports the production of sclerosis of the kidneys by feeding carbonate and acetate of lead in small doses to guinea-pigs and dogs respectively. The condition was associated with a typical lead anemia, but his observations correspond with those of Petroff in that he did not at any time find albumin or casts in the urine. In a more recent article[14] he reports a series of experiments on dogs and rabbits with potassium bichromate. By repeated subcutaneous injections he was able to produce definite foci of interstitial change, though he found that the kidneys had a strong tendency to recuperate even after severe subacute parenchymatous nephritis. In this series of experiments albumin and casts were found in the urine.

Haven Emerson[15] has found that repeated inhalations of ether, alcohol, amyl nitrite and chloroform produced varying degrees of chronic nephritis in dogs, and that in certain cases the manner of death of the animals would indicate that "the condition of the kidneys was probably responsible for their deaths." He also found that intrarenal injections of alcohol, and intramuscular injections of the acetate

11. Ehrlich: Ueber die Zusammenhang von chemischer Constitution und Wirkung. 1898.

12. Levaditi: Experimentelle Untersuchungen über die Necrose der Nierenpapillæ. Arch. internat. de Pharmacod., 1901, viii, 45.

13. Pässler and Heineke: Versuche zur Pathologie der Morbus Brightii. Verhandl. d. deutsch. path. Gesellsch., 1905, ix, 99.

14. Ophüls (W.): Some interesting points in regard to experimental chronic nephritis. Jour. Med. Research, 1908, xviii, 497.

15. Haven (Emerson): An experimental and critical study of the etiology of chronic nephritis. The Archives of Int. Med., 1908, i, 485.

of lead produced chronic lesions in the kidneys, and that simple punctures into the kidney substance were followed by new growth of connective tissue along the line of puncture which, however, did not extend out into the surrounding kidney substance.

Leopold[16] has reported the occurrence of an incipient fibrosis in the kidney of a dog which had been subjected to prolonged administration of sodium chlorid; and recently Siegel[17] has referred to a condition found in a dog thirty days after acute poisoning with uranium nitrate which he describes as a chronic parenchymatous nephritis.

EXPERIMENTAL METHOD.

The animals taken for experiment were well grown guinea-pigs which weighed between 550 and 700 gm. They were fed on carrots and hay, except when under special observation in metabolism cages, when they received a weighed amount of carrots only. A limited number were observed in metabolism cages, from which the total amount of urine was collected and daily tests made. Bacterial growth in the urine was controlled by placing a few drops of chloroform in the receiving vessels. The ordinary clinical tests of the urine were applied, and quantitative estimations of the chlorid and phosphate excretion were made, using Volhard's method for the chlorids, and the uranium nitrate method for the phosphates. The reaction was so alkaline that casts were rarely found in the twenty-four hour specimen. Autopsy was done in every case as soon after death as possible, and, in all cases where the animal was killed, chloroform was used, and the autopsy was done at once. Gross examination was made of all the organs, and histologic examination of the kidneys. Of the various fixing fluids tried, Carnoy's kidney fixative was found to give the best results, although sections fixed with this fluid could not be studied with Sudan III for the presence of fat. The blocks were embedded in paraffin, the sections cut at from four to six microns and mounted with glycerin and egg-albumen. The stains used were hematoxylin and eosin, Weigert's elastic tissue stain, Weigert's fibrin-stain (to demonstrate the hyaline change), and Van Gieson's connective tissue stain. The presence of calcium salts was demonstrated by von Kossa's silver nitrate solution.

16. Leopold: Ueber der Einwirkung von Salzen auf die Nieren (im Tierexperiment). Ztschr. f. klin. Med., 1906, lx, 490.

17. Siegel: Ein Stoffwechselversuch bei Urannephritis am Hunde. Ztschr. f. exper. Path. u. Therap., 1907, iv, 561.

Three series of experiments were made:

1. Eight animals (of which six are reported) were given very frequent subcutaneous injections of 0.25 mg. of uranium nitrate in aqueous solution, from 46 to 87 doses being given in from 77 to 120 days respectively. In this way it was hoped that I might simulate in a certain degree the long-continued mild intoxication which is considered to be an etiological factor in the development of the primary contracted or small red kidney.

2. Seven animals (of which five are reported) were given subcutaneous injections of 2.5 mg. of uranium nitrate at intervals of

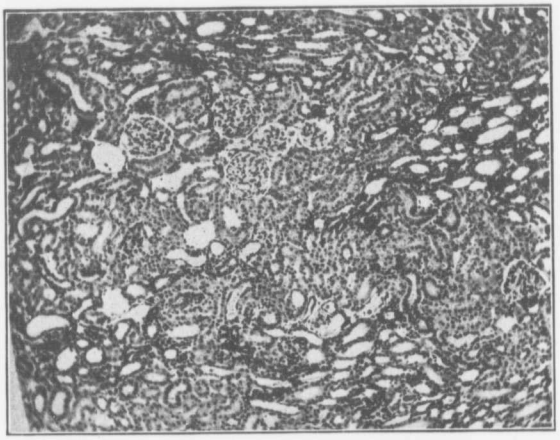

Fig. 3.—Guinea-pig 26: 36 injections of 0.25 mg. in 45 days. Very low power showing the wide distribution of the dilated tubules and the cellular infiltration between them.

from 10 to 30 days, the number of doses and the interval being determined in each case by the reaction of the animal as indicated by the changes in the weight and the urinary picture. The dose was sufficient in each case to produce a definite nephritis, which however was not severe enough to kill the animal. In this way I hoped to be able to show the effect on the kidneys of repeated subacute attacks of nephritis.

3. Fourteen animals were given a single subcutaneous injection of 5 mg. of uranium nitrate, which was sufficient to produce a severe nephritis with extreme loss of weight and almost complete anuria. In two cases death followed within five days, in two cases on the seventh day, and in one case on the eighth day. Of the remaining nine animals two were killed on the twenty-fifth day, two on the one hundred and seventeenth day, one died on the two hundred and twenty-fourth day, two were killed on the two hundred and fortieth day, and two animals were lost. This group (of which eight are reported) will show the condition of the kidneys at different periods after a single attack of acute nephritis.

PROTOCOLS.—SERIES I

PROTOCOL 1—*Guinea-pig 22.*—Eighty-two subcutaneous injections of 0.25 mg. of uranium nitrate were given during one hundred and ten days, after which the animal was killed. The weight decreased from 609 gm. to 455 gm. Albumin was present in the urine for four days following the fortieth dose, but was not observed again.

Autopsy.—The animal was fairly fat and showed no congestion of the peritoneum and no ascites. The kidneys appeared normal in size and color, and on section showed no macroscopic changes. The capsule stripped easily.

MICROSCOPIC EXAMINATION

The majority of the glomeruli show practically no change except that the tufts are dilated and completely fill the capsules. Many of the capsules, however, show definite thickening of the endothelium and of the basement membrane, while others are dilated, forming scattered microscopic cysts in the cortex. A few capsules contain a granular exudate but no desquamated cells. The tufts in nearly all cases show no changes except the dilatation described above, although a few have some thickening of the intervascular connective tissue, and some others show slight fragmentation of the nuclei. The thickened basement membranes of the capsules show definite hyaline degeneration, but there is no hyaline change in the tufts.

The greater number of the convoluted tubules present a fairly normal appearance, although the epithelial cells are somewhat swollen. There are areas, however, where the tubules are distinctly dilated. In these areas the epithelium is flattened and degenerated, in some cases showing merely a narrow band of protoplasm with poorly staining nuclei inside the basement membrane. These tubules are found in the neighborhood of the glomeruli which show changes, and they contain a granular exudate and in many cases some desquamated epithelial cells. In other places the tubules are shrunken and atrophied and show a deposit of pigment. In the medullary rays the ascending limbs of Henle's loops are dilated and contain granular exudate and desquamated cells which show varying degrees of degeneration. The collecting tubules in the pyramids also contain exudate and debris, but otherwise show no change. A few of the tubules contain dense hyaline casts.

There are definite areas of round-cell infiltration in the interstitial tissue, in many cases widely separating the atrophied tubules which in these places show hyaline degeneration of their basement membranes. In a few cases these

areas of round-cell infiltration are found around slightly damaged glomeruli. No changes can be observed in the walls of the blood vessels, and there are no areas of calcification.

PROTOCOL 2—*Guinea-pig 24.*—Fifty-one subcutaneous injections of 0.25 mg. of uranium nitrate were given during sixty-eight days, and the animal died on the sixty-eighth day. The weight decreased from 580 gm. to 415 gm. Albumin was present in the urine after the fourth, fourteenth and twenty-sixth doses for one day each time, after the thirty-second dose for three days, and after the forty-third dose for two days. There was almost complete anuria for the twenty-four hours preceding death, the small amount of urine excreted containing a high percentage of albumin. No convulsions were observed.

Autopsy.—The peritoneum was considerably congested and there was a small amount of clear fluid in the peritoneal cavity. The bladder contained about 5 c.c. of urine, which gave a definite reaction for albumin but contained no casts. The kidneys were rather large and pale, almost gray in color, with some dilated venules over the surface. There was no perinephric fat, and the capsule stripped easily. The cut surface bulged on section and showed some edema. The cortex was paler in color than the medulla.

MICROSCOPIC EXAMINATION

The condition found is similar to but more marked than that described in Protocol 1. There are a few definite areas of fibrosis which seem to replace the medullary rays, and which extend to and correspond with small dimples on the surface. Immediately on either side of these processes the greater part of the parenchymatous change is seen, the appearance of the cortex in other places being practically normal. The glomeruli show changes similar to those described in Protocol 1, but in addition, a few show newly formed elastic tissue fibers around the capsule, external to the thickened, hyaline basement membrane. The cystic formation is more marked and occurs both in the glomeruli and in the convoluted tubules, one cyst being found in the cortex which measured fully 1 mm. in diameter. The ascending limbs of Henle's loops contain greater numbers of desquamated cells but no dense hyaline casts. There are a few small foci of calcium deposit in the cortex and very definite deposit in the medulla.

PROTOCOL 3—*Guinea-pig 25.* (Figs. 1 and 2.)—Forty-six injections of 0.25 mg. were given in seventy-seven days, and the animal died on the seventy-seventh day. The weight dropped from 665 gm. to 410 gm. Albumin was present in the urine for two days after the fifth dose, and for two days before death. There was no noticeable tendency to anuria during the twenty-four hours preceding death, but numerous granular casts were found in the urine at this time.

Autopsy.—The peritoneum was markedly congested and firmly adherent to the abdominal wall in the region where the injections had been made. A very small quantity of fluid was found in the peritoneal cavity. The bladder was full of clear urine which gave a marked reaction for albumin and contained very many granular casts. The stomach and intestines were enormously distended and contained a large quantity of fluid material. The liver, heart, lungs and adrenals showed no gross change. The kidneys were very large, the left being larger than the right, and were dark in color and mottled. The capsule stripped easily. On section there was definite bulging and some edema, and the cortex and medulla were uniformly congested.

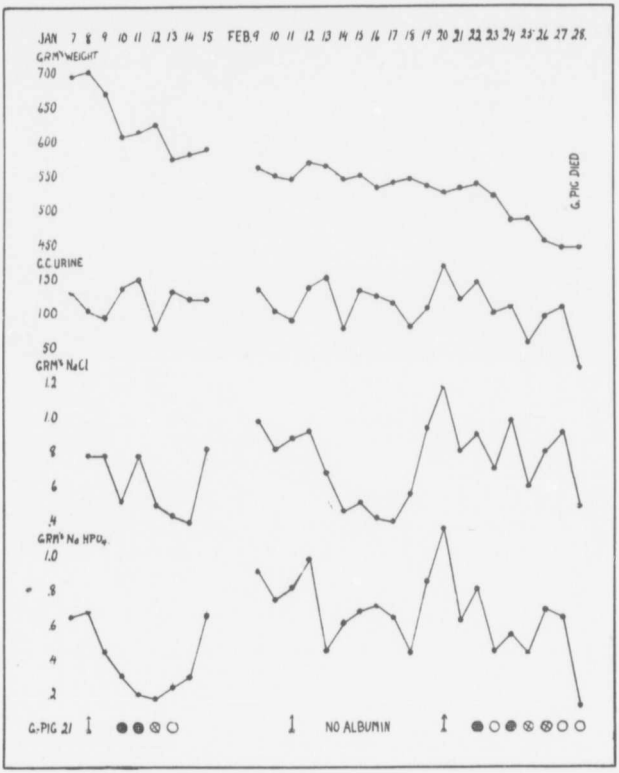

Fig. 4.—Chart showing weight and excretion of urine in Guinea-pig 21. The dose given at each injection was 2.5 mg. The arrows indicate the time at which the injections were given. The shaded circle indicates a large amount of albumin in the urine, the crossed circle indicates a moderate amount, and the plain circle indicates a faint trace. The amount of urine is given in c. c. per twenty-four hours, the amount of chlorids in grams of NaCl, and the amount of phosphates in grams of Na_2HPO_4. The food intake was constant. Note the definite retention of chlorids and phosphates.

MICROSCOPIC EXAMINATION

The greater part of the cortex appears fairly normal, although the glomerular tufts completely fill the capsules, and the cells of the convoluted tubules are swollen and somewhat granular. Some of the glomeruli show thickened capsules with beginning proliferation of the endothelium, and some are surrounded by areas of round-cell infiltration. (Fig. 2.) The greatest change is seen in the medullary rays where the tubules are dilated, their epithelium flattened, and their lumens filled with exudate and desquamated cells. (Fig. 1.) Many of the convoluted tubules in the vicinity show a similar change, and there are areas at and near the cortical margin where the same condition obtains. There are some desquamated cells and exudate in the lumens of the collecting tubules. There are no areas of definite fibrosis, but there are many areas of round-cell infiltration occurring chiefly near the junction of the cortex and medulla, and frequently in close relation to the medium-sized veins. There are no dense hyaline casts in the tubules and very little hyaline degeneration of the basement membranes. A few small patches of calcification are seen in the medulla.

PROTOCOL 4—*Guinea-pig 26.* (Fig. 3.)—Thirty-six injections of 0.25 mg. were given in forty-five days, and the animal died on the forty-fifth day. The weight dropped from 587 gm. to 480 gm. Albumin was present in the urine for one day after the eighth dose and for one day before death. There was almost complete anuria for sixteen hours before death, but no convulsions were observed.

Autopsy.—The peritoneum was definitely congested, and was adherent to the abdominal wall in the region where the injections had been made. There was a considerable amount of clear fluid in the peritoneal cavity. The bladder contained about 7 c.c of clear urine, which gave a definite reaction for albumin and contained granular casts, some epithelial cells, and a few leucocytes. The stomach and intestines were distended and were filled with a fluid material. The heart, lungs, liver and adrenals showed no gross changes. The kidneys were about normal in size and color. The capsule stripped easily and there was some bulging of the cut surface on section.

MICROSCOPIC EXAMINATION

The condition shown is very little different from that described above. There is no definite fibrosis, but there are many areas of round-cell infiltration, and a fair number of glomeruli with thickened capsules which are surrounded with atrophied tubules. There is definite hyaline degeneration of the basement membranes, both of the tubules and of the glomerular capsules, and there is some cyst formation, chiefly of the glomeruli. The epithelium of the tubules is not granular, but many of the tubules are dilated, have flattened epithelium, and contain exudate and desquamated cells in their lumens. (Fig. 3.) Many dense hyaline casts are present in the tubules, but no areas of calcification are seen.

PROTOCOL 5—*Guinea-pig 30.*—Eighty-three injections of 0.25 mg. were given in one hundred and twenty days, and the animal was killed on the one hundred and fifty-seventh day. The weight fell from 540 gm. to 470 gm. during the time the injections were being made, but went up to 615 gm. during the following thirty-seven days. The urine was not observed until after the forty-sixth dose. Albumin was present in the urine for three days after the fifty-first dose, and for four days after the fifty-seventh dose. No casts were found.

Autopsy.—The peritoneum was not congested, there was a very small amount of clear fluid in the peritoneal cavity, and all the organs, including the kidneys, appeared normal on gross examination.

MICROSCOPIC EXAMINATION

There is much less change than in the preceding animals, but there are a number of glomerular cysts which are surrounded by degenerated and atrophied tubules. Some of the glomerular capsules contain exudate, and a few of them have thickened basement membranes which have undergone hyaline degeneration. There is less tubular change than in the preceding cases, but some of the tubules are dilated and have exudate and débris in their lumens. No dense hyaline casts are seen. There are a number of areas of round-cell infiltration scattered through the cortex, but there is no definite fibrosis. There is no new formed elastic tissue, and no deposit of calcium salts.

PROTOCOL 6—*Guinea-pig 36.*—Eighty-seven injections of 0.25 mg. were given during one hundred and eight days, and the animal was killed on the one hundred and eighth day. The weight dropped from 572 gm. to 510 gm. No albumin was observed in the urine.

Autopsy.—There was no congestion of the peritoneum, but a small amount of fluid was found in the peritoneal cavity. The liver, lungs, heart and adrenals showed no gross changes. The kidneys were rather large but normal in color. Immediately beneath the capsule at the lower pole the left kidney contained a large cyst, about the size of a No. 6 shot, which was filled with a clear fluid. No other cysts were seen, and on section the kidney substance appeared quite normal.

MICROSCOPIC EXAMINATION

There is much more change in the kidney substance than one would have expected to find from looking at the gross specimen. There are many areas of round-cell infiltration and many areas of beginning fibrosis which extend to and correspond with definite though microscopic dimples on the surface of the kidney. There are rather numerous microscopic cysts which are derived from both glomeruli and tubules, and which contain a homogeneous exudate. Some of these cysts are surrounded by dense round-cell infiltration, and others, which are larger, have compressed the tubules which lie near them. Many of the cysts show merely a flattened band of epithelium which is degenerated and possesses very poor staining properties, and many of them have a definitely thickened basement membrane. Some of the glomeruli show thickening and hyaline degeneration of the basement membrane, while others contain exudate and desquamated cells. Many of the tubules are dilated and contain exudate and débris, and many of them are atrophied and show hyaline change in their basement membranes. No dense hyaline casts or newly formed elastic fibers are seen. There is no deposit of calcium salts.

PROTOCOLS.—SERIES II

PROTOCOL 1—*Guinea-pig 21.* (Figs. 4, 5 and 6.)—Six subcutaneous injections of 2.5 mg. were given during a period of seventy-seven days. The doses were given on the first, second, twenty-seventh, forty-seventh, sixtieth and seventieth days, and the animal died on the seventy-seventh day. No examination of the urine was made immediately after the first two doses. There was definite albumin excretion for four days after the third dose, and two days later there was a faint trace for one day. Albumin was not observed again until after the sixth dose when it was present for the six days preceding death. There was rather marked irregularity of the chlorid and phosphate excretion in the urine throughout the experiment, with definite retention after the third and fifth doses, in the latter case occurring without any albuminuria. Following the sixth dose there was marked irregularity of the chlorids without definite retention, but there was a very definite phosphate retention. There

was definite loss of weight after the second, third and sixth doses, and the animal dropped from 732 gm. to 455 gm. during the experiment. Figure 4 shows the effects of the third, fifth and sixth doses.

Autopsy.—The animal was rather thin. There was some congestion of the peritoneum but no fluid in the peritoneal cavity. The bladder contained about 5 c.c. of urine which showed a trace of albumin, some leucocytes and many granular casts. The kidneys were about normal in size with little fatty capsule. They were pale in color, slightly mottled, and showed numerous uneven depressions on the surface. On section there was no bulging and no edema, and the cortex was paler and narrower than normal. There was a considerable amount of fluid in the pericardial sac. The other organs appeared normal in the gross except for a large bunch of cysts which were attached to the cystic duct.

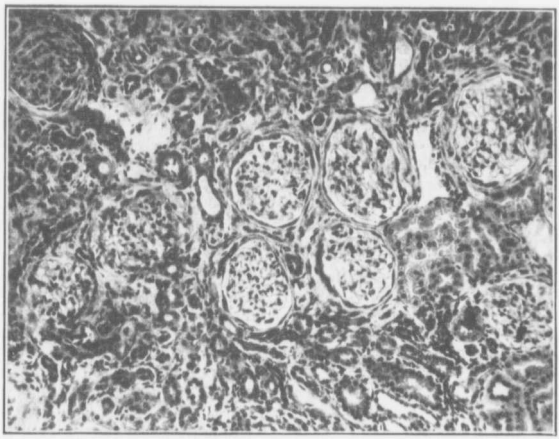

Fig. 5.—Guinea-pig 21: 6 injections of 2.5 mg. during 77 days. Glomeruli with thickened capsules and beginning proliferation of the endothelium, extreme atrophy of the tubules, and marked proliferation of the inter-tubular connective tissue.

MICROSCOPIC EXAMINATION

There is a very definite sclerotic change involving the greater part of the cortex, although it is most marked near the junction of the cortex and medulla. From this deeper part numerous processes pass out towards the margin, and in some places correspond to definite dimples on the kidney surface, though in the majority of cases there is a narrow layer of more or less normal cortex between these processes and the margin. There are a few small cysts scattered throughout the cortex, the majority of which are glomerular in origin. There are very marked degenerative changes in the glomeruli. In most cases there is very little damage to the tufts, but in a few cases there are varying degrees of proliferation

of the connective tissue, and in a few there is complete destruction with hyaline degeneration. The capsules are very much thickened with swollen hyaline basement membranes, and with some proliferation of the endothelium. There are no desquamated cells or exudate in the intracapsular spaces. (Fig. 5.) Surrounding many of the capsules just external to the basement membrane, there is a definite new formation of elastic tissue, which in some cases seems to be continuous with that of the small arteries of the cortex. (Fig. 6.) In most cases there is very extensive connective tissue proliferation around the changed glomeruli, but in some cases the glomeruli seem to be isolated spots of fibrosis which are surrounded by quite normal appearing parenchyma.

The majority of the tubules are involved in the fibrotic changes described above, although at the margin of the cortex, and between the processes of fibrosis which extend to the surface, there are areas of tubules which show practically no change. The damaged tubules are in the main atrophied and compressed, in many cases being represented by bunches of poorly stained nuclei, but some are dilated, having flattened epithelium, and others are distinctly cystic. The dilated tubules contain a granular exudate and desquamated epithelial cells. The basement membranes in many cases are thickened and show hyaline degeneration, but there is no elastic tissue proliferation around the tubules.

The newly formed connective tissue is rich in nuclei throughout, and there are also areas of dense round-cell infiltration. There is a finely granular deposit of calcium salts scattered rather uniformly throughout the fibrotic areas in the cortex. The tubules in the medulla and in the medullary rays are somewhat dilated and contain desquamated cells, exudate, and some dense hyaline casts in their lumens. There is practically no proliferation of connective tissue in the medulla, but there are a few scattered areas of calcium deposit.

Protocol 2—*Guinea-pig 23.* (Fig. 7.)—Six subcutaneous injections of 2.5 mg. were given during a period of seventy days. The doses were given on the first, second, thirty-fifth, fifty-seventh, sixty-third and seventieth days, and the animal died on the seventy-third day. Albumin was present in the urine for four days after the fifth dose, and for the three days after the sixth dose which preceded death. Granular casts were found after the sixth dose. There was a definite increase in the amount of urine excreted during the last two weeks of the experiment, the output for the last eighteen days averaging 123 c.c. per day as compared with 94 c.c. per day for the thirty-one days preceding. The excretion of the salts increased somewhat with the increase in the amount of urine, but the specific gravity for the last eighteen days averaged only 1019 as compared with 1022 for the preceding thirty-one days. There was a definite diminution in chlorids and phosphates after the third, fifth and sixth doses. The animal lost weight after the second, third, fourth and sixth doses, dropping from 680 gm. to 412 gm. during the experiment. The reaction was less marked after every injection than in Guinea-pig 21. Figure 7 shows the reaction after the third, fifth and sixth doses.

Autopsy.—The animal was emaciated, the peritoneum was somewhat congested, and there was a small amount of fluid in the peritoneal cavity. There was a small amount of urine in the bladder which showed some albumin and many granular casts. The kidneys were slightly swollen and mottled, and had no fatty capsule. On section there was some bulging of the cut surface and a slight uniform congestion of both cortex and medulla. The other organs appeared normal on gross examination.

MICROSCOPIC EXAMINATION

There is remarkably little change as compared with that found in Guinea-pig 21. The greater part of the cortex appears normal, although a number of the glomeruli have thickened capsules with some proliferation of the endothelium. There is no exudate and no desquamation of endothelium into the capsular space. There is some increase in the connective tissue of the tufts and some hyaline change in the basement membranes of the capsules, but there is no new formation of elastic tissue. Some of the tubules are dilated, having flattened epithelium and containing exudate and débris in their lumens. There are a few casts in the loops of Henle, and a few areas of round-cell infiltration which are chiefly in the region of the larger veins. There are no definite areas of fibrosis, and there is no deposit of calcium salts.

PROTOCOL 3—*Guinea-pig 28.*—Seven subcutaneous injections of 2.5 mg. were given during a period of ninety-four days. The injections were given on the third, eighteenth, thirty-third, forty-sixth, sixty-ninth, eighty-sixth and ninety-fifth days. The urine was observed for a short time only in the early part of the experiment. Albuminuria was present after the fifth and seventh doses, and the chlorid and phosphate excretion was very irregular at this time. The animal lost weight after all except the third injection. The weight dropped from 580 gm. to 420 gm. during the period of injection, but increased to 610 gm. during the following forty-two days.

Autopsy.—The animal was very fat, the peritoneum was not congested, and there was no fluid in the peritoneal cavity. There was a large amount of perinephric fat, and the kidneys were rather small and mottled. In places the surface was distinctly dimpled, and in these areas the capsule was adherent. On section the cortex appeared paler than the medulla, and was narrower than normal. The small amount of urine in the bladder contained neither albumin nor casts. The other organs appeared normal on gross examination.

MICROSCOPIC EXAMINATION

There are many patches of fibrosis scattered through the cortex, and some of these extend to the surface and correspond to the dimples on it. The fibrotic areas are not confined to the inner zone of the cortex, but are irregularly scattered through it. There are a good many small cysts, most of which are glomerular in origin. The majority of the glomeruli have thickened capsules, some have proliferation of the endothelium, and others show some fibrosis of the tufts. There is no exudate into the intracapsular spaces. There is hyaline degeneration of the thickened basement membranes of the capsules, and in some cases there is a new formation of elastic tissue. In the areas of fibrosis there are bunches of glomeruli which show the changes described, and scattered through the more normal appearing cortex there are many single glomeruli which show a similar condition. The convoluted tubules do not show much atrophy except in the sclerotic areas, but around the changed glomeruli there are some dilated tubules with flattened epithelium which have exudate and desquamated cells in their lumens. The collecting tubules and the loops of Henle also contain exudate and a few casts. There is some hyaline degeneration of the basement membranes of the tubules in the fibrotic areas, but there is no new formation of elastic tissue. There is some deposit of calcium salts in the medulla, but none in the cortex.

PROTOCOL 4—*Guinea-pig 34.*—Six subcutaneous injections of 2.5 gm. were given during a period of eighty-three days. The doses were given on the second, twenty-second, thirty-sixth, forty-fifth and eighty-third days, and the animal died

on the ninety-seventh day. The urine was observed for a short time only. Albumin was found after the fifth and sixth doses, and the excretion of salts was very irregular. The animal lost weight after the second, third, fourth, fifth and sixth doses, and continued to lose weight during the fourteen days which preceded death. The total loss of weight was from 547 gm. to 250 gm. The reaction after each injection was very severe, the animal becoming acutely ill each time.

Autopsy.—The animal was very thin and the peritoneum was somewhat congested, but there was no fluid in the peritoneal cavity. A very small amount of urine in the bladder gave a definite test for albumin, but unfortunately an examination for casts was not made. The kidneys were very small and distinctly granular, but had no fatty capsule. The capsule was rather firmly adherent to the surface of the kidney. On section the kidney substance was firm, the cortex thin and pale, and the medulla dark. The other organs appeared normal on gross examination.

MICROSCOPIC EXAMINATION

There is very definite proliferation of the connective tissue throughout the cortex with marked destruction of the glomeruli and tubules. The glomeruli show marked thickening of the capsules with hyaline degeneration of the basement membranes and new formation of elastic tissue. There is definite increase in the connective tissue of the tufts, which appear much more dense than in any of the other cases. Many of the tubules are atrophied and shrunken; others are dilated, having flattened epithelium, and contain exudate in their lumens. There is very little desquamation of the epithelial cells. There are very dense hyaline casts in the loops of Henle and in the smaller collecting tubules. The connective tissue proliferation is very widely distributed and extends well down into the medulla, where it separates the tubules in a rather marked degree. The connective tissue is sparsely nucleated and there are no areas of round-cell infiltration. There is very marked calcium deposit in both medulla and cortex, the calcification being so marked as quickly to destroy the edge of the knife when cutting sections.

PROTOCOL 5—*Guinea-pig 37.*—Six subcutaneous injections of 2.5 mg. were given during a period of one hundred and eleven days, and the animal was killed on the one hundred and thirty-fourth day. Albumin was present in the urine after the first, fifth and sixth doses, the only times at which observations were made. There was definite retention of salts after the first dose, and a very irregular excretion after the fifth and sixth doses. There was a definite increase in the amount of urine excreted during the latter part of the experiment. During the first nine days the average output was 89 c.c. per day, with an average specific gravity of 1020, and during the last fifteen days the average excretion was 109 c.c. per day with an average specific gravity of 1018. The animal lost weight after the first, third and sixth doses, dropping from 495 gm. to 405 gm. during the experiment.

Autopsy.—A small amount of fluid was found in the peritoneal cavity, but the bladder was practically empty. The kidneys were small, somewhat pale in color, and were slightly granular on the surface. On section there was no bulging and no edema, the consistency was somewhat more resistant than normal, and the cortex was rather narrow.

MICROSCOPIC EXAMINATION

There is a very marked fibrosis of the cortex of the kidney with extensive destruction of the tubules and glomeruli. The new formed connective tissue is very rich in cells, and is quite diffuse. It is most marked in the inner zone of the

cortex, but there are many processes which extend to the surface of the kidney, and which correspond to the dimples on the surface. The majority of the glomeruli show thickening of their capsules, proliferation of their endothelium, and hyaline degeneration of their basement membranes. Many of the tufts are condensed and fibrous, and a few show more or less hyaline change. There are many small glomerular cysts, some of which contain exudate, and there is new-formed elastic tissue around some of the glomeruli. In some areas the tubules appear fairly normal, but in many places they are dilated, have flattened epithelium, and contain exudate in their lumens, and in other places they are more or less completely atrophied, in some places being represented by bunches of poorly staining nuclei. The loops of Henle and the collecting tubules contain exudate and débris, and many of the loops contain dense, fragmented, hyaline casts. There is some

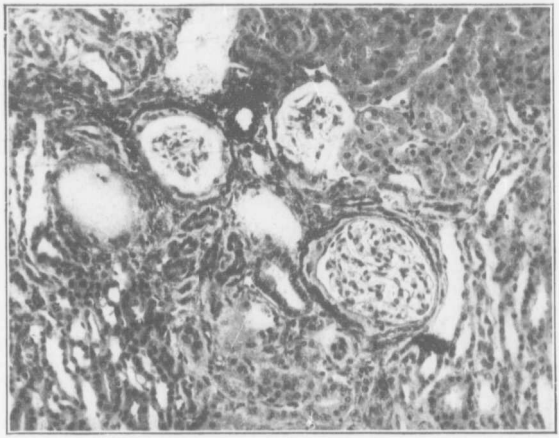

Fig. 6.—Guinea-pig 21: New-formed elastic tissue around slightly damaged glomeruli. It is situated outside the thickened basement membrane and is closely associated with the small artery. (Weigert's elastic tissue stain.)

hyaline degeneration of the basement membranes of the shrunken tubules, but there is no newly formed elastic tissue around them. There are many areas of calcification in the cortex, and some of the medulla.

PROTOCOLS.—SERIES III

PROTOCOL 1—*Guinea-pig 32.*—A single injection of 5 mg. was given subcutaneously, and the animal died four days later. The loss in weight was about 25 gm.

Autopsy.—The peritoneum was slightly congested, but there was no fluid in the peritoneal cavity. The kidneys were dark in color, and larger and less firm

than normal. The capsule stripped easily. On section there was definite bulging of the cut surface and some edema, and both cortex and medulla were distinctly congested.

MICROSCOPIC EXAMINATION

There is very marked congestion of the kidneys and an extreme degree of damage to the convoluted tubules, but there is very little change in the glomeruli. The only indication of interstitial change is seen in a few areas of round-cell infiltration around some of the small veins. The glomeruli are practically normal in appearance, although there is slight congestion of the tufts, and in a few cases some exudate into the intracapsular space. There is no thickening of the capsule, and no desquamation or thickening of the capsular endothelium. The convoluted tubules are practically all enormously degenerated; many of them are dilated with their epithelium flattened and their lumens filled with exudate, and many are practically denuded of their epithelium. Those tubules where the damage is less severe show cloudy degeneration of the cells and almost complete obliteration of their lumens. The ascending limbs of Henle's loops are greatly dilated and contain exudate and desquamated cells, and the smaller collecting tubules also contain considerable débris. A few hyaline casts are seen in the tubules. There is no deposit of calcium salts.

PROTOCOL 2—*Guinea-pig 48.*—A single injection of 5 mg. was given subcutaneously, and the animal died five days later. There was a very heavy excretion of albumin in the urine for the four days preceding death, and there was also some copper-reducing substance which gave a very marked "sugar" reaction, for three days preceding death. The animal dropped in weight from 570 gm. to 295 gm.

Autopsy.—The peritoneum was congested, but there was no fluid in the peritoneal cavity. The kidneys in the gross and microscopically were practically the same as those described in Protocol 1 of this series.

PROTOCOL 3—*Guinea-pig 16.*—A single injection of 5 mg. was given subcutaneously, and the animal died on the eighth day. The weight dropped from 530 gm. to 380 gm. No analysis of the urine was made.

Autopsy.—There was marked congestion of the peritoneum, but no fluid in the peritoneal cavity. The kidneys were swollen, and on section showed some edema and some bulging of the cut surface. The capsule stripped easily. The other organs showed no gross changes.

MICROSCOPIC EXAMINATION

There is very definite congestion of the arterioles and of the glomeruli. There are numerous, rather large cysts scattered throughout the cortex, many of which are tubular in origin though some are glomerular. Many of the glomeruli are unchanged except for the congestion of the tufts, but some have exudate into the glomerular capsule, a few show some proliferation of the capsular endothelium, and an occasional one shows some thickening of the basement membrane of the capsule. The majority of the tubules show very little change except for some cloudy swelling of the epithelium. Some of the tubules are dilated, having flattened epithelium, and containing exudate and débris in their lumens, and some of them show calcium deposit in the cells of the tubules and in the desquamated cells in their lumens. The loops of Henle and the smaller collecting tubules have some exudate in their lumens, and a few contain hyaline casts. There are a few areas of round-cell infiltration and a good deal of calcium deposit in the cortex. There is no new formation of elastic tissue.

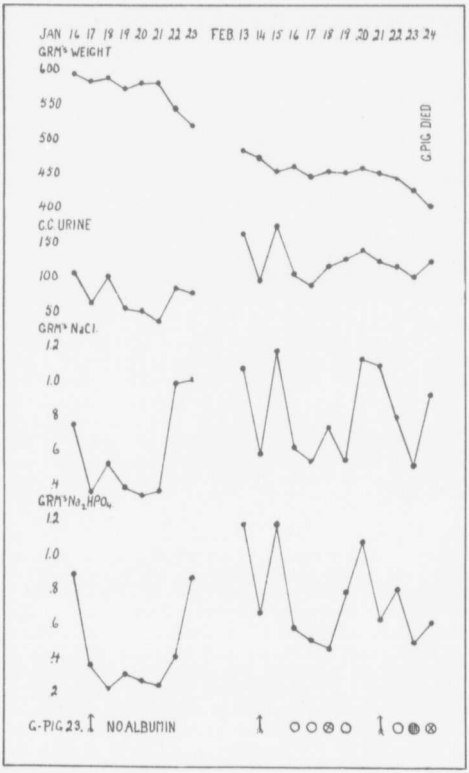

Fig. 7.—Chart showing weight and excretion of urine in Guinea-pig 23. Note the definite retention of chlorids and phosphates and the increased amount of urine excreted in the latter part. For full explanation, see legend of Figure 4.

PROTOCOL 4—*Guinea-pig 19.* (Fig. 8).—A single injection of 5 mg. was given subcutaneously, and the animal was killed on the twenty-fifth day. The weight dropped from 545 gm. to 360 gm. during the first fifteen days, but increased to 405 gm. during the following ten days. No analysis of the urine was made.

Autopsy.—There was no congestion of the peritoneum, but there was a small amount of fluid in both the pleural and peritoneal cavities. Neither kidney had any fatty capsule. The right kidney was slightly swollen and very pale though mottled in color. On section there was some bulging of the cut surface which had a peculiar grayish color. There was a fine striation of calcareous infarction along the outer margin of the medulla. The left kidney was much smaller than the right, and was chalky white in color. On section the whole cut surface of the kidney was uniformly white and the pelvis of the kidney was filled with a milky white fluid which contained a few leucocytes, and numerous small white globules which dissolved with effervescence in dilute acid. The other organs showed no gross changes.

MICROSCOPIC EXAMINATION

The type of change found is very similar in both kidneys, but has progressed much farther in the left than in the right. The right kidney shows an outer zone of cortex (about one-half the width of the cortex), in which there is very little change in either the glomeruli or the tubules. The inner half of the cortex, however, shows very definite change. The capsules of the glomeruli are very much dilated, forming small cysts. There is no thickening of the basement membrane of the glomeruli, and except in a few cases there is no proliferation of the endothelium. The tufts are slightly shrunken but not collapsed, and they show no increase in their connective tissue elements. The intracapsular space is dilated to about five-thirds its original size, and in most cases does not contain any exudate. (Fig. 8.).

The tubules of the inner zone show varying degrees of change. Some are dilated, having flattened epithelium and containing exudate in their lumens, while others are collapsed and show a moderate though not an advanced degree of atrophy of the epithelium. In these areas the tubules are separated by a recent cellular proliferation of the interstitial tissue. There are very many fragmented hyaline casts in the ascending limbs of Henle's loops, and there is exudate and desquamated cells in some of the collecting tubules. There are a few small patches of degenerated tubules in the outer zone of the cortex which show some increase in their cellular elements, and which correspond to microscopic dimples on the surface of the kidney. There are many large areas of round-cell infiltration in the inner zone of the cortex which are mostly situated in the neighborhood of the larger veins. There is no hyaline change in the basement membranes of the glomerular capsules or of the tubules, and there is no new formation of elastic tissue.

The left kidney, which was the smaller, presents a much more advanced degree of change. The condition described in the inner zone of cortex in the right kidney, is found throughout the whole cortex of the left kidney, so that there is practically no normal appearing parenchyma left. Almost all the glomerular capsules are dilated and show no thickening of their basement membranes, although a very few have a slight proliferation of the endothelial cells. Practically all the tubules are degenerated, some being dilated and containing

exudate and desquamated cells, while others are collapsed and more or less completely atrophied. Many of the loops of Henle contain casts, some of which are hard and fragmented, and many of the collecting tubules contain exudate and desquamated cells. There is definite increase in the intertubular connective tissue, and there are many areas of round-cell infiltration. There is no hyaline degeneration of the basement membranes of either the tubules or the glomeruli, and there is no new formation of elastic tissue. There are several areas where calcium granules are seen inside the cells of the degenerating epithelium.

PROTOCOL 5—*Guinea-pig 33.* (Fig. 9.)—A single injection of 5 mg. was given subcutaneously, and the animal was killed in one hundred and seventeen days. No analysis of the urine was made. The weight dropped from 520 gm. to 387 gm. during the first ten days, but subsequently increased to 505 gm.

Autopsy.—The peritoneum was not congested, but there was a small amount of fluid in the peritoneal cavity. Both kidneys were small, the left being smaller than the right, and both were red in color, distinctly granular, and surrounded by a well marked fatty capsule. The true capsule was adherent in both kidneys, and on section the cortex appeared narrower than normal. There was no bulging of the cut surface and no edema.

MICROSCOPIC EXAMINATION

The picture presented is that of an advanced interstitial nephritis. There is very great destruction of the renal tissue, marked proliferation of the connective tissue, extensive cyst formation, and definite irregularity of the surface of the kidney. (Fig. 9.) The most marked degree of change is in the inner zone of the cortex, but large areas of fibrosis extend to the surface and correspond to the depressions on it. The glomeruli show very marked change throughout. In the inner zone of the cortex many of them are cystic, varying in size of from two to three times their normal diameters. Some of them are empty, some show remnants of their more or less compressed tufts, and others contain exudate and desquamated cells. Their capsules show considerable thickening, notwithstanding the fact that they are so greatly distended. The glomeruli in the peripheral zone are about normal in size, but there are practically none which do not show more or less definite change. The tufts completely fill the intracapsular spaces, and show some fibrosis and some fragmentation of the nuclei, but no hyaline change. The capsules are very much thickened, there is definite hyaline degeneration of the basement membranes, and in many cases marked proliferation of the endothelial layer. Many of the glomeruli and cysts are surrounded by newly formed elastic tissue which is situated immediately outside the thickened basement membrane.

The convoluted tubules are also very much damaged. There are a few small areas where the tubules have a fairly normal appearance, but even here there is an increased number of intertubular nuclei, and in many cases the tubule is completely isolated by a thin but definite layer of connective tissue which surrounds it. The vast majority of the tubules are atrophied and shrunken, being represented by a ring of small misshapen nuclei. In some cases there is a small lumen which contains a granular exudate and desquamated cells, but there is no dilatation of the convoluted tubules. Many of the degenerated cells contain a fine granulation of calcium deposit, and the basement membranes of the atrophied tubules show marked hyaline degeneration. Many of the ascending limbs of Henle's loops are dilated to several times their normal diameter, and are lined by very much flattened and degenerated epithelium. Many of these tubules are empty, but some of them contain exudate and

desquamated cells, and others contain dense, fragmented, hyaline casts. Many of the collecting tubules contain exudate and débris.

There is very marked proliferation of a richly cellular connective tissue especially in the cortex where it extends to the surface and corresponds to the depressions which are found on it. There are very definite areas of round-cell infiltration, some of which are associated with the larger veins. There are some areas of calcium deposit in the medulla.

PROTOCOL 6—*Guinea-pig 35.* (Fig. 10.)—A single injection of 5 mg. was given subcutaneously, and the animal was killed on the one-hundred-and-seventeenth day. The weight dropped from 540 gm. to 460 gm. during the first eight days, but had increased to 530 gm. at the time of death. No analysis of the urine was made.

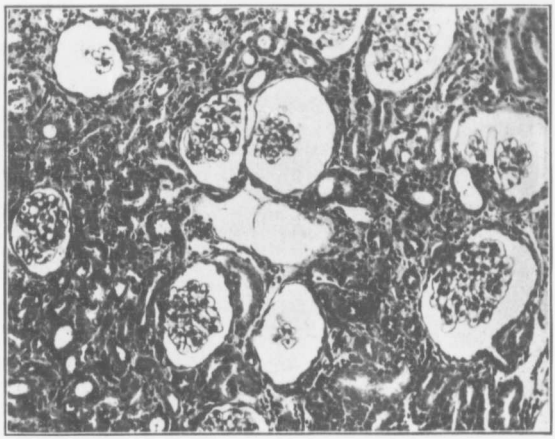

Fig. 8.—Guinea-pig 19: A single injection of 5 mg. The animal was killed on the twenty-fifth day. Cystic dilatation of the glomerular capsules, beginning proliferation of the interstitial tissue, and beginning atrophy of the tubules.

Autopsy.—The animal was fat and there was no congestion of the peritoneum. There was a small amount of fluid in the peritoneal cavity. The kidneys were somewhat swollen and rather pale. The left kidney had a large cyst, about the size of a BB shot, situated in the upper pole at the inner margin of the cortex. The other organs showed no changes visible on gross examination.

MICROSCOPIC EXAMINATION

The type of change closely resembles that described in Protocol 5, although it is much less marked in degree. There is rather extensive fibrotic change in the inner zone of the cortex, but only in a few places does the fibrosis extend

to the surface of the kidney and cause a depression on it. There are much larger areas of normal appearing cortex, but here too, there are many tubules which are completely surrounded by thin processes of connective tissue. Many of the glomeruli in these areas show little if any change, but some have thickened capsules with proliferation of the endothelial layer. In the inner zone, however, the condition closely resembles that found in Guinea-pig 33. There is definite cyst formation, extreme atrophy of the tubules, marked proliferation of the connective tissue, and thickening of the glomerular capsules. (Fig. 10.) There is no marked dilatation of the loops of Henle, but many of them are filled with exudate and desquamated cells, and others contain dense fragmented casts. Many of the collecting tubules contain exudate and débris. There is marked hyaline degeneration of the basement membranes of the thickened capsules and atrophied tubules, and there is definite new formation of elastic tissue around some of the glomeruli. There is some granular calcium deposit in the degenerating epithelium, and some scattered areas of calcification in the medulla.

PROTOCOL 7—*Guinea-pig 47.*—A single injection of 5 mg. was given subcutaneously, and the animal was killed on the two-hundred-and-fortieth day. The weight dropped from 580 gm. to 335 gm. in the first ten days, but had increased to 475 gm. on the seventeenth day. It was not observed at time of death. There was a steady decrease in the amount of urine during the first twelve days until there was almost complete anuria. Albumin appeared in the urine on the third day after the injection and was still present on the seventeenth day. The "sugar" reaction appeared on the third day and lasted four days. No further examinations of the urine were made during the life of the animal, but after death there was a small amount of urine in the bladder which contained a few leucocytes, some epithelium and a few granular casts.

Autopsy.—The animal was very fat, the peritoneum was slightly congested, and there was a small amount of fluid in the peritoneal cavity. The kidneys were not swollen but were somewhat pale and rather mottled. The capsule stripped easily. On section there was no bulging of the cut surface and no edema. The other organs appeared normal on gross examination.

MICROSCOPIC EXAMINATION

The cortex of the kidney shows many small cysts which are mostly glomerular in origin. Many of the glomeruli have thickened capsules with hyaline degeneration of their basement membranes, and a few are surrounded by newly formed elastic tissue. Many of the convoluted tubules are dilated, having flattened epithelium and containing exudate and desquamated cells in their lumens, and many of the loops of Henle are blocked with dense, fragmented, hyaline casts. There is no marked diffuse new growth of the intertubular connective tissue, and there are no areas of round-cell infiltration. There are a few areas of calcium deposit occurring chiefly in the medulla.

PROTOCOL 8—*Guinea-pig 44.*—A single injection of 5 mg. was given subcutaneously, and the animal died on the two-hundred-and-twenty-fourth day. The weight dropped from 510 gm. to 440 gm. during the first six days, and the animal weighed 385 gm. at time of death. There was no decrease in the amount of urine excreted after the injection, but there was definite albuminuria commencing on the second day and persisting throughout the seven days during which observations were made. The "sugar" reaction appeared on the second day and lasted for four days. The animal gained weight rapidly after the first ten days, and remained in apparently good health until within a few days before death.

Autopsy.—The peritoneum was slightly congested, and there was a small amount of fluid in the peritoneal cavity. The kidneys were somewhat swollen and mottled, and the capsule stripped easily. The cortex appeared normal. There was no urine in the bladder.

MICROSCOPIC EXAMINATION

The appearance is very much the same as that described in Protocol 7, although there is not quite so much glomerular change. There are many normal glomeruli, but there are also many glomerular cysts, and many glomeruli with thickened capsules, hyaline basement membranes, and new formed elastic tissue. Some of the convoluted tubules are dilated and have exudate and débris in their lumens. Many of the loops of Henle are blocked by dense, fragmented casts, and many of the collecting tubules contain exudate and desquamated cells. There are some small areas of fibrosis which are rich in nuclei, and which in some places correspond to dimples on the surface of the kidneys, but there is no diffuse proliferation of the intertubular connective tissue. There is some deposit of calcium salts in the medulla.

DISCUSSION

Before discussing the significance of the foregoing experiments it may be well to refer briefly to an objection which will undoubtedly be raised. It is well known that small animals are subject to spontaneous kidney lesions, and that conditions are occasionally found which more or less closely resemble that of chronic interstitial nephritis in man. In view of this fact one must be careful that a condition which may be coincident with, be not interpreted as resultant from the administration of some renal irritant.

In our series of experiments sixteen animals which died as a result of acute poisoning with uranium nitrate and four which were killed as normal controls were carefully examined for evidences of chronic lesions. Of these twenty animals not one showed any condition which at all resembled those described above (excepting, of course, the two acute cases described in Protocols 1 and 2 of Series III). In one animal which succumbed to acute poisoning a single glomerulus was found which showed slight thickening of the endothelium, but in no case was there any thickening of the capsule or hyaline degeneration of the basement membrane, and in none of the kidneys of the normal controls was there any appearance of round-cell infiltration.

On the other hand, in Series I, in which eight animals were subjected to experiment, one was killed after a few injections but before anything was found, one died from acute poisoning with sodium chlorid and so could not be included in the series, and the other six are reported. And in Guinea-pig 31 which died from sodium chlorid poisoning, the kidneys showed glomerular and tubular lesions which were undoubtedly not due to the terminal insult.

In Series II, in which seven animals were injected, five are reported. One animal, No. 29, received seven injections but showed practically no reaction at any time, and, when killed after one hundred and thirty-four days, showed no definite or characteristic lesions. Another animal, No. 38, was killed by an overdose of sodium chlorid and so could not be included in the series. Animals 21, 28, and 34 which showed the most marked reaction during life showed also the most severe kidney lesions after death.

In Series III, in which fourteen animals were injected, two died within five days and are reported as examples of acute poisoning, two

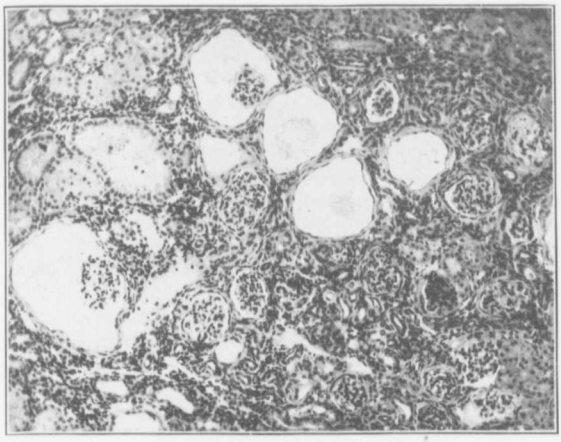

Fig. 9.—Guinea-pig 33: A single injection of 5 mg. The animal was killed in 117 days. Low power showing marked cystic dilatation of the glomerular capsules, and advanced fibrosis with atrophy of the tubules and thickening of the glomerular capsules. The low power will give some idea of the wide distribution of the fibrotic change.

were lost, and six of the others are reported. In only one instance, Animal 20, was there a complete absence of kidney lesion; the other three all showed definite changes which in kind, though not in degree, resembled those described in the cases reported.

Now when it is considered that in the examination of twenty animals which had not been subjected to experiment not one was found

which showed any chronic lesions; that out of twenty-seven animals subjected to treatment only two failed to show some definite and characteristic change; and that in all three series those animals which reacted most actively during life showed the most marked degrees of change after death; I think I am justified in assuming that the lesions which were found were due to the administration of Uranium Nitrate.

From 1885 to 1889 Chittenden with Hutchinson and Lambert[18] investigated the pharmacology of uranium nitrate, and noted that in dogs it produced acute parenchymatous nephritis which was associated with albuminuria and later with glycosuria. It was not until after Richter's report[6] in 1904, however, that especial attention was paid to the action of this drug as a renal irritant. Since that time it has been extensively used in the production of experimental nephritis, but the attention of investigators seems to have been almost exclusively focused upon a study of the process of edema formation which is associated with acute poisoning with this drug. Indeed if I exclude Siegel's[17] reference to an incipient chronic change in the kidneys of a single dog, I have been unable to find any reference in the literature to the use of uranium nitrate in attempts to produce chronic nephritis in animals.

Schlayer and Hedinger[19] and Takayasu[20] have investigated the reaction of normal and damaged kidneys to stimuli of various kinds, and they found that kidneys damaged by uranium nitrate, together with those damaged by potassium bichromate and corrosive sublimate, constitute a group which they designate as "tubular," in contrast to those damaged by cantharidin and arsenic, which they designate as "vascular." Histologically they found that in the tubular group of nephritis the epithelium of the tubuli contorti suffered the greatest damage, while that of the straight and collecting tubules remained intact. They also found slight injury to the Malpighian corpuscles, but they state that this was more marked in those kidneys which were poisoned by potassium bichromate. Takayasu[20] says that the tubular change is the characteristic feature of the tubular group, and that the glomeruli show definite changes only comparatively late. He says that even in cases of severe functional disturbance there is very slight damage to the glomeruli. Desquamation of the capsular endothelium

18. Chittenden and others: Cited from Tylecote: Pharmacology and therapeutics of uranium. Med. Chron., Manchester, 1904, 379.

19. Schlayer and Hedinger: Experimentelle Untersuchungen über toxische Nephritis. Deutsch. Arch. f. klin. Med., 1907, xc, 1.

20. Takayasu: Ueber der Beziehungen zwischen anatomischen Glomerulusveränderungen und Nierenfunktion bei experimentellen Nephritiden. Deutsch. Arch. f. klin. Med., 1907, xcii, 127.

is seen in only a few cases, and in very slight degree. The blood content of the glomeruli is normal, and there is no change in the tufts. There is no increase in the number of the nuclei in the glomeruli although in some cases they show distinct enlargement and loss of staining power. He points out, however, that these latter changes are especially seen in the bichromate kidneys, and that they are present in a much less degree in the uranium and corrosive sublimate kidneys, not more than 20 per cent of the nuclei of the uranium kidneys being so changed.

Christian[21] has recently reported the occurrence of small round or oval, homogeneous, hyaline droplets, which he found in the glomerular tufts of thirteen out of twenty-six rabbits examined, and of these thirteen, eleven were animals which had been poisoned by uranium nitrate. The droplets appear in the wall of the capillaries which make up the glomerular tufts, and do not occur in the lumen of the capillaries, or in the space between the tuft and the capsule of the glomerulus, or in the endothelium of Bowman's capsule.

Protocols 1 and 2 in Series III describe the histologic picture which I found in the acute intoxications. The convoluted tubules and the ascending limbs of Henle's loops showed early and marked degeneration, and contained exudate and desquamated cells. The epithelium of the ascending limbs of Henle seemed to be the earliest cells affected, and there was marked degeneration and desquamation of these cells while those of the convoluted tubules were still relatively intact. The collecting tubules contained exudate and débris, but showed no degeneration of the epithelium. There was some congestion of the glomeruli, and slight exudate into the intracapsular space in a few cases, but we did not observe the nuclear changes described by Takayasu or the hyaline droplets described by Christian. There was some change in the interstitial tissue as was shown by the areas of round-cell infiltration in those cases which survived for a few days, but there was no thickening of the glomerular capsules, and no visible increase in the intertubular connective tissue. The feature to which I would draw especial attention in the acute poisoning is the extreme degree of change in the tubules, and the very insignificant amount of change in the glomerular tufts and capsules.

In Series I, in which repeated small doses were given, the results obtained, while perhaps not very marked, were sufficiently uniform and striking to justify further observations over a longer period of time. It is unnecessary to repeat in detail the picture described in the proto-

21. Christian: A glomerular lesion of experimental nephritis. Boston Med. and Surg. Jour., 1908, clix, 8.

cols, but I may briefly draw attention to the following facts. Five of the six animals reported showed albuminuria at some time during the course of the experiment, and of the three which died as a result of the injections, two showed very definite anuria before death. At autopsy five were found to have a certain amount of ascites, and of the four cases where urine was present in the bladder, in one the urine contained albumin but no casts, in two it contained albumin and granular casts, and in the fourth it was clear. In three cases the kidneys were about normal in size and color, and in the other three they were somewhat swollen. In two of the latter three cases the color

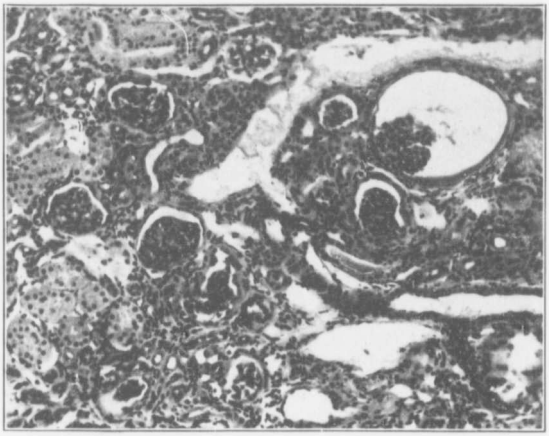

Fig. 10.—Guinea-pig 35: A single injection of 5 mg. The animal was killed on the 117th day. Cystic dilatation of the glomerular capsule and moderate fibrosis with atrophy of the tubules, and some thickening of some of the glomerular capsules.

was darker than normal and mottled, while in the other it was rather pale. In every case the capsule stripped easily.

The microscopic picture (Figs. 1, 2, and 3) was very similar in all the kidneys although the degree of change present was more marked in some. The glomerular tufts were dilated and completely filled the capsule in every case. Many of the glomeruli were apparently normal, but in each case there were some capsules which showed thickening and hyaline degeneration of the basement membrane, and

some proliferation of the endothelial layer. There was distinct cystic formation of the glomerular capsules in five cases, and new formation of elastic tissue around some of the glomeruli in one. There was more or less degeneration of the tubules in all, with atrophy of the epithelium and dilatation of the lumens in which were exudate and desquamated cells. The greater number of the tubules appeared normal, and the areas of degeneration which were present were more or less closely associated with the areas in which the glomeruli were most severely damaged. There were distinct areas of round-cell infiltration in every case, occurring chiefly near the larger veins at the inner margin of the cortex, but in two cases there was definite infiltration in the region of the damaged glomeruli. The greatest amount of tubular change was found associated with these areas of round-cell infiltration. In four of the cases there was no definite increase in the intertubular connective tissue, but in the other two there was definite fibrosis in the medullary rays which extended to the surface and corresponded with distinct though microscopic dimples on it. There was deposit of calcium salts in two cases.

These results would indicate that long-continued, mild intoxication with uranium nitrate will produce a beginning fibrosis in the kidneys. It would seem that the condition is progressive, and it is interesting to note that the animal which showed the greatest amount of fibrosis in the kidneys had received the greatest number of injections. The picture rather closely resembles that described by Weigert in subgroup 1 of the "chronic hemorrhagic with heart hypertrophy," and although I am unable to say that the animals' hearts were hypertrophied, I believe that the kidneys of this series may be classed under that heading.

In Series II the results obtained were just as constant and much more striking. Each injection was sufficiently strong to produce a definite subacute attack of nephritis from which the animal usually recovered. A glance at the charts, Figures 4 and 7, will give some idea of the results of each injection. In nearly all cases there was a definite albuminuria with more or less retention of the chlorids and phosphates. There was definite loss of weight after almost every injection. In two cases there was definite polyuria towards the end of the experiment, and similar comparison could not be made in two of the remaining cases because the animals had not been kept in the metabolism cages for a sufficiently long time. As stated above, I was unable to find casts in the twenty-four-hour samples of urine except on rare occasions, but in two of the four cases in which there was urine in the bladder after death, numerous granular casts were found.

and in one of the other cases in which the urine was rich in albumin, a microscopic examination was unfortunately not made. In only one case was the urine obtained in the bladder free from both albumin and casts. At autopsy only two of the animals had any fluid in the peritoneal cavity, but in four there were distinct macroscopic dimples in the cortex of the kidneys to which the capsule was adherent, and in one of these the kidneys were small, pale and distinctly granular.

The microscopic picture (Figs. 5 and 6) showed marked proliferation of the interstitial connective tissue with distinct dimpling of the surface of the kidney and marked tubular and glomerular destruction. In one case the new growth of fibrous tissue seemed to be more marked in the inner zone of the cortex, but in the other four it was quite diffuse, and in two cases it extended to the intertubular connective tissue in the medulla. The most striking feature was the advanced degree of glomerular change, a fact that is all the more remarkable when we remember that in the acute intoxication there was comparatively little involvement. The glomerular changes included thickening of the capsules with hyaline degeneration of the basement membranes and proliferation of the endothelium in every case, new formation of elastic tissue around some of the glomerular capsules in four cases, cystic formation in three cases, rather marked proliferation of the connective tissue of the tufts in two cases, and complete hyaline degeneration of a few of the tufts in one case. The convoluted tubules were severely damaged in four cases, and moderately so in one. There was marked atrophy and practical obliteration of many of the tubules, and dilatation with degeneration of the epithelium in many others. The dilated lumens of the latter contained exudate and desquamated cells. Many of the ascending limbs of Henle's loops were dilated and contained exudate and débris, and some of them were blocked by dense hyaline casts. The collecting tubules showed little damage to their epithelium but contained exudate and desquamated cells. The newformed connective tissue was fairly rich in cells and there was round-cell infiltration in one case. The newly formed elastic tissue was found only around the damaged glomeruli outside the thickened basement membrane, and in some cases it seemed to be continuous with the elastic tissue in the walls of the small arteries. There was some deposit of calcium salts in the medulla in two cases, and rich deposit in both medulla and cortex in two cases.

The results obtained in this group show very definitely that following a series of subacute attacks of nephritis such as is produced by uranium nitrate there is marked and permanent damage to the kidneys. The marked increase in the interstitial tissue, the extreme

degree of tubular and glomerular change, and the occurrence of polyuria towards the end of the experiment, show a decided resemblance to the conditions found in chronic interstitial nephritis in man. Although I cannot say that the blood pressure was high, nor that there was definite hypertrophy of the left ventricle of the heart, yet the analogy to those conditions described by Weigert as the "most chronic" of the "chronic hemorrhagic with heart hypertrophy," is, I believe, sufficiently strong to justify a classification under that heading. Indeed in the one case (Guinea-pig 21), one might almost be justified in classing the kidneys as true secondary contracted or granular atrophic kidneys.

In Series III I can not draw comparisons as I have in Series I and II, because of the difference of the duration of the experiments, and because I have no complete records of the urine excretion during the experiments. There was, however, this in common, viz., that in every case the animal was so ill for several days after the injection that it was doubtful whether or not he would recover. I have already referred to protocols 1 and 2 in my discussion of the microscopic picture in acute poisoning by uranium nitrate. Guinea-pig 16 (Protocol 3) and Guinea-pig 15 (which also died on the seventh day, but which is not reported in detail), show already a beginning proliferation of the endothelium of the glomerular capsules and numerous areas of round-cell infiltration, although the most striking feature is that even this early there is definite cystic formation of the glomeruli and also of the tubules. In Guinea-pig 19 (Protocol 4), Fig. 8, there is a very marked cystic formation which is chiefly glomerular in origin, and in the left kidney, which is the least damaged, it is seen that the condition apparently commences in the inner zone of the cortex. This is of interest because we shall see that the two most severely damaged kidneys of this group also show this characteristic. There is rather marked though recent new-formed connective tissue between the tubules, and the fact that the condition is progressive is evidenced by the areas of round-cell infiltration. The glomeruli show beginning proliferation of the endothelium in a few cases, but there is no definite thickening of the capsule, and no hyaline degeneration of the basement membrane. This kidney is interesting in that it undoubtedly shows an early stage of the condition to which we shall refer later, and that it also shows how early we may find well-marked proliferation of the interstitial tissue, and advanced cystic dilatation of the glomerular capsules. Guinea-pigs 33 and 35 (Protocols 5 and 6), Figs. 9 and 10, furnish examples of the more advanced changes. Both show advanced fibrosis of the kidneys with marked tubular and glomerular changes, with the formation of cysts, hyaline degeneration of the basement mem-

branes, and new formation of elastic tissue around some of the glomeruli. In Guinea-pig 35 the change is almost entirely confined to the inner zone of the cortex, although there are some processes which extend to the surface and correspond to distinct depressions on it. In Guinea-pig 33, however, the condition is much more general, and although there is greater cystic degeneration of the glomeruli in the inner zone of the cortex, yet almost all the cortex has undergone extensive fibrotic change. In Protocols 7 and 8 we have changes which, although slight as compared with those in Guinea-pig 33, are nevertheless definitely of the nature of chronic lesions. There is cyst formation and rather extensive glomerular thickening, but there is no general increase in the intertubular connective tissue.

A review of this series cannot fail to show that the lesions described in Protocols 4, 5, and 6 must be closely related; and the rather marked cystic change described in Protocol 3 would indicate that this must also be included. The results show definitely that following a single severe attack of parenchymatous nephritis, such as is produced by uranium nitrate, there may be severe and permanent damage to the kidneys which may go on so far as to result in the production of a granular, atrophic kidney. Protocols 5 and 6 show this most distinctly, and I think that Protocol 4 may also be included as showing an earlier stage of what must undoubtedly have resulted in a condition similar to that described in Protocol 5. The findings in Protocols 7 and 8 would indicate that in other cases, although there may be a certain number of permanent chronic lesions produced in the kidneys, there must be a remarkable tendency to recuperation even after exceedingly severe attacks of acute parenchymatous nephritis.

The most striking feature in these three series is the extreme degree of glomerular change which is found in the kidneys that show chronic lesions. As I noted above, this is all the more remarkable when we remember the insignificant amount of damage to the glomeruli which is found in the acute intoxication with this drug. In Series I there is the possibility that the condition is due to the long-continued mild irritation of the repeated small doses, but in Series II and III this cannot be the explanation. In these two series the change is undoubtedly secondary to the damage done by the acute poisoning, and in this it bears out Senator's teaching that the whole tube system is damaged as a result of injury to any part of the system. Ponfick[22] believes that such a condition may be the result of a passive process in which the

22. Ponfick: Ueber Morbus Brightii (Referat). Verhandl. d. deutsch. path. Gesellsch., 1905, ix, 49.

glomerulus becomes atrophied through disuse because of a prolonged plugging of the tubule by a dense hyaline cast. Müller,[4] on the other hand, suggests that it may be a non-progressive fibrosis due to scar formation, in which the scar is the result of the injury to the parenchyma by the primary acute attack. In my series, however, there is a quite constant feature which it may be well to mention, as it may tend to support Senator's theory[2] that the secondary contracted kidney is the result of a progressive process because of some damage to the vascular system. In all the advanced cases of Series III, in four of the five cases in Series II, and in one case in Series I, new-formed elastic tissue fibers were found around some of the damaged glomeruli. Melnikow-Ruswedenkow[23] has described this condition in a series of diseased human kidneys in which he found the elastic tissue around the glomerular capsules, outside the thickened basement membrane. (In my cases it was invariably in this situation that the new-formed elastic tissue was found. See Fig. 6. In some instances in which it was found in a scar near the surface of the kidney, the elastic fibers seemed to be derived from the elastic tissue in the capsule of the kidney, but in all other cases the origin was in the walls of the blood vessels. He says definitely that its occurrence depends on the vascular system, that it may be present in either general or local circulatory disturbances, and that Bowman's capsule is the most frequent seat because it stands in such intimate relation to the walls of the arterial branches. Hohenemser[24] demonstrated the presence of newly formed elastic fibers in the kidney tissue in seventeen cases of arteriosclerotic and chronic inflammatory kidneys. He says that it occurs only in the region of, and in closest dependence upon the blood vessels, and that in common the amount of elastic tissue present stands in direct relation to the amount of shrinkage of the organ affected. Now in view of these explanations as to the origin of the new formed elastic tissue in the kidney (which, by the way, are all the references on the subject that I was able to find in the literature at my disposal), if we consider also that in Series III the lesions appeared to commence and to be more severe in the inner zone of the cortex which is near to the large blood vessels, and that in all my cases of acute poisoning which showed round-cell infiltration, and in many of the cases in Series I, the areas of round-cell infiltration seemed to be in the neighborhood of the larger vessels, it may be that uranium nitrate causes some damage

23. Melnikow-Ruswedenkow: Histologische Untersuchungen über die elastische Gewebe in normalen und in pathologische veränderten Organen. Beitr. z. path. Anat. u. g. allg. Path. (Ziegler's), xxvi, 564.

24. Hohenemser: Ueber der Vorkommen von elastischen Fasern bei cirrhotischen Prozessen d. Lebers und Niere. Virchow's Arch. f. path. Anat., cxl, 192.

to the vascular system which so far we have been unable to demonstrate, and it may be that it is this vascular disturbance which is responsible for the secondary glomerular and tubular destruction. It is interesting to note in this connection that Heineke and Meyerstein[25] concluded from their observations on acute uranium poisoning that there must be some vascular change because of the occurrence of severe edema in cases in which there was very little renal damage.[26]

It is not my intention to try to explain the results obtained. I feel that perhaps I should apologize for suggesting a classification for the types described in Series I and II, but I make that classification merely to emphasize the very close resemblance which exists between the lesions produced by uranium nitrate poisoning and those which are found in the various types of subacute and chronic diffuse nephritis in man. I believe I have demonstrated beyond question that it is possible to produce in animals conditions which closely resemble those found in subacute and chronic interstitial nephritis in man, and that during the development of these conditions I have in certain cases been able to demonstrate characteristic urinary findings.

CONCLUSIONS

1. By the use of uranium nitrate it is possible to produce in animals lesions which closely resemble those found in subacute and chronic diffuse nephritis in man.

2. A long-continued mild intoxication with this drug will lead to the production of a "subchronic" (Weigert) interstitial nephritis which would appear to be progressive.

3. A series of several attacks of subacute uranium nephritis will lead to extensive fibrotic changes in the kidney which in some cases goes on to the stage of a granular atrophy. In a certain number of cases this condition is associated with the condition of polyuria.

4. A single severe attack of acute parenchymatous nephritis may be followed after some weeks by a more or less severe fibrosis, which in some cases goes on to extreme granular atrophy. In other cases, however, the recuperative power of the kidney is such that only a moderate fibrosis results.

---

25. Heineke and Meyerstein: Experimentelle Untersuchungen über die Hydrops bei Nierenkrankheiten. Deutsch. Arch. f. klin. Med., 1907, xc, 101.

26. It may be of interest to mention here that in one guinea-pig, No. 26, and in one rabbit (out of two injected), distinct plaques of intimal and medial thickening were found in the aorta near the aortic valves. The number is too small to draw any conclusions as the lesions may have been spontaneous, but careful examination will be made of the aorta in all our future cases.

Reprinted from The Archives of Internal Medicine, June, 1909

AMERICAN MEDICAL ASSOCIATION, ONE HUNDRED AND THREE DEARBORN AVENUE
CHICAGO