GLOMERULONEPHRITIS PRODUCED IN DOGS BY SPECIFIC ANTISERA

II. PATHOLOGIC SEQUENCES FOLLOWING THE INJECTION OF RABBIT ANTIDOG-PLACENTA SERUM OR RABBIT ANTIDOG-KIDNEY SERUM*

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PLATES 93 TO 101

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Glomerulonephritis in the rat and rabbit produced by specific antikidney serum has been extensively investigated and the lesions of the acute and chronic disease have been reported (1-5). It also has been shown that rats develop a progressive glomerulonephritis when injected with rabbit antirat-placenta serum and that if the animals are pregnant, abortion ensues (6).

Studies on nephritis in the dog have been carried out by Wilson and Oliver (7) who established the specificity of rabbit antidog-kidney serum in producing nephritis. These workers sacrificed their animals 10 to 22 days after injection, allowing only one to survive for 119 days. Fauts, Corcoran, and Page (8) studied the effect of repeated injections of antidog-kidney serum in dogs while attempting to produce exacerbation of the nephritis.

In the preceding paper (9) the natural history of nephritis in the dog resulting from a single injection of nephrotoxic serum has been described. The present report describes the lesions produced in these pregnant and non-pregnant dogs following injection of either rabbit antidog-placenta or rabbit antidog-kidney serum. Acute and chronic phases of the nephritis have been studied over a period of 10 months.

Materials and Methods

Eleven dogs, 7 beagles and 4 mongrels, were injected with 2.0 to 3.0 ml. per kilo of rabbit antidog-placenta serum. Four of the dogs were in the middle third and one in the last third

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TABLE I
Lesions in Dogs Injected Intravenously with Rabbit Antidog–Placenta Serum

<table>
<thead>
<tr>
<th>Dog</th>
<th>Time after injection (days)</th>
<th>Biopsy</th>
<th>Glomerulonephritis</th>
<th>Severity</th>
<th>Time after last injection (days)</th>
<th>Autopsy</th>
<th>Glomerulonephritis</th>
<th>Severity</th>
</tr>
</thead>
<tbody>
<tr>
<td>M4</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>Uremia</td>
<td>8*</td>
<td>Kidneys mottled, smooth surface with punctate hemorrhages. Hemorrhages in stomach and cecum. Necrotizing arteritis of stomach</td>
<td>Acute</td>
<td>++++</td>
</tr>
<tr>
<td>M7</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>Uremia</td>
<td>9</td>
<td>Kidneys large. Punctate hemorrhages of kidney, stomach, and cecum. Necrotizing arteriolitis of stomach and kidneys</td>
<td>Acute</td>
<td>++++</td>
</tr>
<tr>
<td>A39</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>Uremia</td>
<td>11*</td>
<td>Kidneys, punctate hemorrhages. Focal necrosis of lymphoid follicles of spleen with arteriolitis of spleen. Pulmonary edema</td>
<td>Acute</td>
<td>++++</td>
</tr>
<tr>
<td>M3</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>Uremia</td>
<td>30</td>
<td>Kidneys pale, finely pitted surface. Acute interstitial pancreatitis not related to blood vessels</td>
<td>Acute</td>
<td>++++</td>
</tr>
<tr>
<td>M6</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>45</td>
<td>Kidneys and organs normal in the gross</td>
<td>None</td>
<td>0</td>
</tr>
<tr>
<td>N1</td>
<td>-</td>
<td>-</td>
<td>Subacute</td>
<td>30</td>
<td>120</td>
<td>Kidneys finely pitted</td>
<td>Chronic</td>
<td>+</td>
</tr>
<tr>
<td>N2</td>
<td>30</td>
<td>Subacute</td>
<td>+++</td>
<td>180</td>
<td>Kidneys large, pale, irregular corticomedullary striations, finely pitted surface</td>
<td>Chronic</td>
<td>++++</td>
<td></td>
</tr>
<tr>
<td>N3</td>
<td>30</td>
<td>Subacute</td>
<td>+</td>
<td>180</td>
<td>Kidneys slightly contracted. Finely pitted surface</td>
<td>Chronic</td>
<td>++</td>
<td></td>
</tr>
<tr>
<td>N4</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>210</td>
<td>Kidneys finely pitted, corticomedullary striations indistinct</td>
<td>Chronic</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td>N5</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>210</td>
<td>Kidneys and organs normal in the gross</td>
<td>None</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>A14</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>315</td>
<td>Kidneys finely pitted, corticomedullary line broad</td>
<td>Chronic</td>
<td>+</td>
<td></td>
</tr>
</tbody>
</table>

* Died. 1 Killed in fight.

of pregnancy. All animals in this group died or were sacrificed by an intravenous administration of sodium pentobarbital 8 to 315 days following injection of antisera.

Five dogs, 3 beagles of known age and 2 mongrels of unknown age, were injected with 2.5 to 4.0 ml per kilo of rabbit antidog-kidney serum. None of these animals was pregnant. They died or were killed with anesthesia from 15 to 91 days after the last injection.

Six dogs, 3 beagles and 3 mongrels, were injected with 3 to 4 ml per kilo of normal rabbit
serum. Of the 3 gravid animals in this group, one was in the first third and 2 in the last third of their pregnancies. They were sacrificed from 11 to 189 days after the last injection.

The sera were prepared and administered according to the methods described in the preceding paper (9), wherein pertinent data are recorded in Tables I, II, and III.

Autopsies were performed on all the injected dogs, and on their issue: the fetuses, the still born, or the viable pups which subsequently died. Tissues were fixed in 10 per cent formalin and in Zenker's solution. In addition to hematoxylin and eosin, azan carmine, phosphotungstic acid hematoxylin, elastic Van Gieson, periodic acid–Schiff (PAS), and Oil Red 0 stains were employed as necessary to demonstrate special features of the lesions.

Tables I, II, and III summarize the pathologic findings in the animals receiving antiplacenta, antikidney, or normal rabbit serum. More complete details of the course of the disease and autopsy findings illustrative of the various phases of the disease are given in the appendix.

**RESULTS**

Typical lesions of glomerulonephritis developed in all but 2 animals receiving antiplacenta serum and in all animals receiving antikidney serum. The kidney lesions were similar with each of the nephrotoxic sera employed. It is to

<table>
<thead>
<tr>
<th>Dog</th>
<th>Time after injection</th>
<th>Biopsy</th>
<th>Severity</th>
<th>Autopsy</th>
<th>Glomerulonephritis</th>
<th>Severity</th>
</tr>
</thead>
<tbody>
<tr>
<td>A56</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>Uremia 30*</td>
<td>Kidneys large, smooth, pale, mottled with pinpoint hemorrhages. Purpuric areas in lungs. CPC liver and spleen. No vascular lesions</td>
<td>Acute</td>
</tr>
<tr>
<td>M18</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>70</td>
<td>Kidneys and organs normal in the gross</td>
<td>Chronic</td>
</tr>
<tr>
<td>M17</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>91</td>
<td>Kidneys and organs normal in the gross</td>
<td>Chronic</td>
</tr>
</tbody>
</table>

* Died.  
† Chronic passive congestion.
be emphasized that the following description of the lesions is a composite account and that the kidney lesion beyond the acute stage varied as did the course of the disease in the animal.

Since all animals which died in the acute phase were in uremia, the renal damage was severe. The kidneys were pale, swollen, and mottled, and the surfaces were studded with hemorrhages. The parenchyma bulged over the cut capsule and the cortical striations were blurred. Microscopically, the lesions appeared to be predominantly exudative in character. Almost every glomerulus was affected, the damage varying from edema of the basement membranes to complete necrosis. Red blood cells and fibrin were present in the capsular spaces. Fibrin and hyaline thrombi occluded the capillaries in partially necrotic glomerular tufts. The basement membranes that were not necrotic were thickened and ill-defined (Figs. 1 and 2).

Within the capillary walls was a hyaline deposit that contained lipid and PAS-positive material. Bizarre giant cells, apparently resulting from coalescence of epithelial cells, were encountered (Fig. 3).

In the acute phase the epithelial cells of Bowman’s capsule were desquamated and those that remained attached to the capsular basement membrane were swollen and contained large hyperchromic nuclei. As early as the 8th and 9th days following the injection of antiserum proliferation of both glomerular and

### TABLE III

*Lesions in Dogs Injected Intravenously with Normal Rabbit Serum*

<table>
<thead>
<tr>
<th>Dog</th>
<th>Sacrificial Time after last injection</th>
<th>Autopsy</th>
<th>Glomerulonephritis</th>
<th>Severity</th>
</tr>
</thead>
<tbody>
<tr>
<td>M15</td>
<td>11 days</td>
<td>Kidneys normal in the gross, Pneumonia</td>
<td>None</td>
<td>0</td>
</tr>
<tr>
<td>M14</td>
<td>42 days</td>
<td>Kidneys and organs normal in the gross</td>
<td>Thickened glomerular basement membranes</td>
<td>±</td>
</tr>
<tr>
<td>M12</td>
<td>84 days</td>
<td>Kidneys and organs normal in the gross</td>
<td>None</td>
<td>0</td>
</tr>
<tr>
<td>41</td>
<td>133 days</td>
<td>Small infarct of kidney, healed. ?Healed arteritis of vasa vasorum and single artery of kidney</td>
<td>Thickened glomerular basement membranes</td>
<td>±</td>
</tr>
<tr>
<td>B3</td>
<td>189 days</td>
<td>Kidneys and organs normal in the gross</td>
<td>None</td>
<td>0</td>
</tr>
<tr>
<td>B2</td>
<td>189 days</td>
<td>Kidneys and organs normal in the gross</td>
<td>None</td>
<td>0</td>
</tr>
</tbody>
</table>

*No biopsies were performed on these animals.*
capsular epithelial cells and adhesions between tuft and capsule were apparent. The interstitial tissue was edematous. A material which appeared to be hyaline in the hematoxylin and eosin preparations and which stained intensely with periodic acid–Schiff stain was often present in the widened periglomerular spaces. Lymphocytes, plasma cells, histiocytes, and occasional polymorphonuclear cells were seen frequently about the glomerular capsule and blood vessels and were scattered diffusely throughout the interstitial tissue. Many of the glomerular capillaries were widely dilated. The damage to the tubules was less severe than that to the glomeruli. The tubular basement membranes appeared normal, being intact, thin, and sharply defined, with occasional wrinkles due to compression by the edematous interstitial tissue. The lumens of the tubules were filled with red blood cells and hyaline, cellular, granular, and hemosiderin casts. The epithelium was frequently desquamated, swollen, or in various stages of disintegration. Hyaline droplet degeneration and hemosiderin granules were present within the epithelial cells. These changes in the epithelium of the convoluted tubules often led to occlusion of the lumen. The calyces and pelves were normal.

Three dogs surviving this period in fairly good health were subjected to biopsy of the kidney on the 30th day. Basement membranes of the glomeruli in this subacute phase were thicker, more compact, and more definitely outlined than in the acute phase. Fewer thrombi were present in the capillaries which often appeared widely dilated and empty. Only occasional red blood cells were noted in Bowman’s spaces although fibrin was still present. Proliferation of the capsular and glomerular epithelium had progressed so that there were numerous adhesions between the two and early crescent formation. Giant cells in the glomeruli were prominent. No arterial lesions were seen. The tubular changes reflected the widespread glomerular damage. Many tubules were atrophic while others were dilated and contained hyaline, granular, and occasional red blood cell casts. Some of the epithelial cells of the convoluted tubules were swollen and granular, their appearance resembling the acute stage. In tubules containing casts, the epithelial cells were flattened and devoid of periodic acid–Schiff-positive material. Mitotic figures indicated attempts at repair of epithelial cells. In contrast to the acute phase the basement membranes of the tubules appeared irregularly thickened. The interstitial tissue was infiltrated with scattered lymphocytes and histiocytes. An occasional wandering cell contained hemosiderin pigment. These cellular elements tended to have a perivascular distribution. A fine interstitial scarring occurred as a result of atrophy of groups of tubules (Fig. 4).

Animals which were sacrificed or died from 70 days to 315 days after injection were considered to be in the chronic phase of their disease. The kidneys usually were not reduced in size but were finely pitted. The cortical striations were slightly blurred but the height of the cortex did not appear reduced. Micro-
GLOMERULONEPHRITIS PRODUCED BY SPECIFIC ANTISERA. II

Scopically, there was a tendency for the lesion to undergo transition from an inflammatory to a reparative state. The basement membrane of the glomeruli was thicker and frequently showed lipid deposits in the capillary walls. Similarly, Bowman’s capsule was thickened by fibrous tissue and by hyaline deposits forming dense adhesions to the glomerular tufts. This material was sudanophilic and periodic acid-Schiff-positive. Some glomeruli were obliterated by fibrous tissue and hyaline deposits. Occasional granular precipitate was present in the space of Bowman.

In one of the 3 animals (N23, Table II) in which biopsy specimens were obtained 2 to 5 months before autopsy, progression of the renal lesions was demonstrable (Figs. 4 and 5). In the remaining two (N2, N3) there seemed to be definite healing of the tubular lesion while the glomerular lesion remained about as severe (Figs. 6 and 7). The dilated convoluted tubules with various types of casts were replaced by scar tissue and atrophic tubules with thickened basement membranes. The relatively few casts which were seen were hyaline and were most numerous in the collecting ducts. Despite the repair of the tubular damage the interstitial tissue did not seem appreciably increased over that seen in the subacute phase. The amount, character, and location of the cellular infiltration remained the same. Dog A14 (Table I) had the longest survival period (315 days). The typical appearance of the mild lesion is illustrated in Fig. 8.

Vascular lesions were encountered in the 2 groups injected with nephrotoxic serum. One of the 4 animals, M7, receiving rabbit antidog-placenta serum and dying in uremia in the acute phase, had a necrotizing arteritis and arteriolitis of the kidneys (Fig. 2). This animal and M4 had a necrotizing arteritis of the stomach. A third animal (A39) had arteriolitis of the spleen as well as of the kidney (Fig. 1) while another (M3) had acute interstitial pancreatitis but this did not appear to be related to the arterial tree. None of the animals killed or dying beyond the 30th day had lesions in organs other than the kidney which were considered related to the renal disease.

One of the two dogs (A26, Table II) which received rabbit antidog-kidney serum and which was sacrificed during the acute phase when it had serum disease and elevated blood urea nitrogen (see Table I, accompanying paper) had a necrotizing arteritis of the stomach as well as fibrinoid degeneration of an arteriole of the kidney (Figs. 9 and 3). Another animal (N23) of this series which died in uremia on the 90th day had buccal ulcers and a necrotizing arteritis and arteriolitis of the kidneys and stomach.

There were questionable renal lesions in 2 of the 6 animals injected with normal rabbit serum (M14, 41, Table III). These lesions consisted of a generalized thickening of the glomerular basement membrane (Fig. 10). No casts or scarring were present. One medium sized artery in one section of the kidney of dog 41 showed an interrupted internal elastica and slight disruption and scar-
ring of the media. There was also a small healed infarct of the kidney. The only other similar arterial lesion occurred in a vas vasorum of a section of the aorta in the same animal (Fig. 11) and possibly represented the vestige of a healed arteritis, since this animal was the only one of the group to develop frank serum sickness on the 10th day after receiving rabbit serum. It continued to show proteinuria until killed 19 weeks after injection. The other dog (M14), which showed mild kidney lesions of the same type was a mongrel of unknown age, though so gray about the muzzle as to be called “Grandma.” The course in this animal was uneventful for 6 weeks after injection at which time she was killed with anesthesia. Proteinuria had occurred on only two occasions. The thickened glomerular basement membrane was probably due to age since the basement membranes of dog glomeruli tend to thicken with age (10).

DISCUSSION

The kidneys of dogs in the uremic phase of acute nephritis induced by antidog placenta or antidog kidney serum were found to be enlarged and studded with pinpoint hemorrhages. The cortex was swollen and the cortical striations were indistinct, simulating the appearance of human kidneys in the acute phase of glomerulonephritis. The kidneys of dog N23 (Table II) which died in uremia 90 days following injection were also enlarged, although this animal was arbitrarily placed in the group having chronic nephritis. However, as reported elsewhere (11), a dog which died in uremia 3 years and 8 months after the injection of antiplacenta serum had pale, greatly contracted, and granular kidneys producing an anatomical counterpart of kidneys in the chronic phase of human glomerulonephritis. Therefore, the kidneys of the dogs in this series, classified in the chronic phase, may not have been reduced in size because of the relatively short duration of the illness.

The extra renal lesions seen in the small group of dogs dying in uremia presented certain similarities to and differences from those observed in man. In common was the frequent occurrence of buccal lesions. Enteritis was not encountered but severe necrotizing arteritis and arteriolitis resulting in ulceration were found in the stomach. Fibrinous pericarditis has not yet been noted nor have the hyaline membranes and relatively acellular exudate characteristic of uremic pneumonia in man been seen. Pulmonary edema and petechial hemorrhages, however, have been observed. Uremic frost was not noted at necropsy but the characteristic ammoniacal odor of the body was present.

In general the cytotoxic nephritis was characterized by severe necrotizing and exudative lesions in the initial phases. The subsequent phases seemed to represent an attempt to repair the initial damage and were characterized by proliferation of fibrous tissue, thickening of the basement membranes, and scarring of the renal parenchyma. Although in the chronic phase the glomerular lesions were diffuse, no fresh exudative lesions were seen. This suggests that
the initial trauma determined the course of the disease and that there was no continued or repeated injury by the agent. Only one animal (N23) of 8 considered to be in the chronic phase had a progressively downhill course terminating in uremia. Two other dogs showed kidney lesions which had not progressed in severity in the 5 months intervening between biopsy and autopsy.

Comparison of the chronic lesions in dogs and man is unwarranted since many factors known to influence the human disease are as yet unknown in the dog. For example the duration of the disease may not be comparable; the hypertension and accelerated arteriosclerosis and arteriolosclerosis often present in the human disease did not appear in the dogs. These superimposed burdens probably accentuate the kidney lesion in man.

It will be noted that 2 of the 11 dogs receiving rabbit antidog-placenta serum had no demonstrable renal lesions at autopsy (Table I). Injection of the maximal dose of serum (3 ml. per kilo) produced proteinuria for 1 month in dog M6 and for 5 months in dog N5. The urine was normal for 2 weeks and 6 weeks respectively before autopsy. Nitrogen retention was absent in both animals. The absence of glomerular lesions in these animals suggests a course parallel to that of acute glomerulonephritis in man, wherein restoration of the kidney to normal is thought to occur in the majority of cases.

Two of the 5 dogs (M18 and M17, Table II) receiving rabbit antidog-kidney serum in smaller amounts (2.7 and 2.5 ml. per kilo) developed proteinuria, with casts and red blood cells in the urinary sediment, and nitrogen retention. The urinary abnormalities were slight and the blood urea nitrogen was normal when the animals were killed. Autopsy showed only minimal renal lesions which were focal rather than diffuse. The glomerular basement membranes were thickened; hemosiderin pigment was seen in the epithelium and a few casts were present in the tubules.

It is impossible to say whether this minimal renal lesion (M18 and M17), represents an anatomic effect of the injection of foreign serum or whether it should be considered an indication of healed nephritis. No vascular lesions were present but the animals were sacrificed 70 and 91 days after injection so it is possible that mild vascular lesions or cellular infiltration in the vessels characteristic of foreign serum reactions might have disappeared. On the other hand, both animals had transiently elevated blood urea nitrogen values and persistent though mild proteinuria. The relation, if any, that this lesion bears to human glomerulonephritis is unknown. Literature on the histologic picture of healed human glomerular nephritis is understandably lacking.

It is possible that the vascular lesions represent a hypersensitive reaction to foreign serum (12). This is probably true in dog 41, which received normal rabbit serum and developed frank serum sickness on the 10th day after injection. It is also possible that the vascular lesions in all the animals dying or sacrificed in the acute phase of the disease may be attributed to hypersensitivity. On the other hand, they may represent a manifestation of uremia.
The failure of pregnancy to affect the manifest course of the disease has been described in the preceding paper (9). The fetuses and placentas of 3 dogs (A39, M4, M3) in acute uremia in the first, second, and last thirds of pregnancy respectively were normal. Autopsies were performed on the stillborn pups and on pups dying in the neonatal period. Beyond the usual maceration of the stillborn fetuses no abnormalities were noted in either group. No kidney lesions were present. Placental sites were examined in all cases and no abnormalities were apparent.

Several investigators (13–15), have indicated that the antigen which stimulates the production of nephrotoxic antibody is to be found in the glomerulus, probably in the basement membrane. Immunologic studies carried out in relation to the present investigation (9) and other observations made by Pressman (16) and by Cruickshank and Hill (17) establish the presence of at least one antigen common to placenta and kidney. These findings provide a possible explanation for the fact that the lesions produced by rabbit antidog–kidney serum and rabbit antidog–placenta serum are indistinguishable and that the glomerular basement membrane is the tissue most seriously and permanently damaged.

**SUMMARY**

The intravenous injection of rabbit antidog–placenta or antidog–kidney serum produced immediate evidence of glomerulonephritis in dogs. The lesions produced by the two agents were indistinguishable.

The glomerulonephritis so induced may terminate in death within 8 days, may progress to a chronic form or may heal. The acute phase of the nephritis was usually associated with severe exudative and necrotizing renal lesions, while the pathologic sequences in surviving dogs were essentially reparative. However, 1 dog of 8 in the chronic phase developed a progressive chronic nephritis and died in uremia.

**APPENDIX**

A39.—Female beagle, 3 weeks pregnant, 33 months old, weighing 11.4 kilos when injected intravenously with a total of 3.0 ml. per kilo of rabbit antidog–placenta serum Jan. 19 to 24, and Jan. 26, 1953. Injections spaced over 1 week because of shock-like reaction to antiserum.

Jan. 27 to Day of Death, Feb. 6.—Proteinuria, 0.86 to 2.88 gm. per cent, with many types of casts and red cells in the urinary sediment. Urea nitrogen retention first noted on Jan. 27th and persisted throughout the period of observation (maximum of 374 mg. per cent on Feb. 3). The dog became hypertensive (maximum 160/110 on Jan. 30) and anemic (lowest red blood cell count 3.48 million cells per c. mm. on Feb. 5). The anemia might have been associated in part with pregnancy. Three determinations of serum protein partition during this period showed reversed albumin globulin ratios. Blood cholesterol was 312 mg. per cent on Feb. 3. From Jan. 28th on, animal appeared weak, refused food, and vomited. Clyses of saline and glucose were given from Jan. 29th until the animal died on Feb. 6, 1953. A bloody vaginal discharge was noted terminally.
Autopsy, Feb. 6, 1953.—The body was that of a pregnant female beagle weighing 10.1 kilos. No free fluid was present in the chest or abdomen. The heart (weight 90 gm.) and aorta were normal. The lungs were congested and contained a large amount of frothy edema fluid. The stomach contained 50 cc. of dark brown fluid. The mucosa of the prepyloric region was congested. The duodenum and jejunum each contained 2 small punched out mucosal erosions. The rectal mucosa was congested.

Both kidneys were enlarged. The right weighed 39 gm.; the left, 41 gm. The surface was smooth but mottled, and pinpoint hemorrhages were visible in the cortex. The corticomedullary zone appeared broad and irregular.

Ureas was distended with 7 macerated fetuses about 2 inches long. The placentas were partially detached. All other organs including the brain were normal.

Microscopic.—

Kidneys showed the most severe acute lesions encountered in the series. Many glomeruli were completely necrotic, others partially so. Rupture of the basement membranes had resulted in exudation of fibrinoid material in the periglomerular spaces. Within the glomeruli there were cellular debris and occasional giant cells. The arterioles were normal but about the glomeruli and throughout the interstitial tissue there was a diffuse infiltration of lymphocytes, plasma cells, histiocytes, and polymorphonuclear cells. Tubular damage was less severe than the glomerular damage. The epithelium was swollen and contained hyaline droplets and hemosiderin granules. Red blood cells often distended the tubular lumens. In periodic acid-Schiff preparations the basement membrane of the tubules was occasionally wrinkled but not thickened giving an appearance of compression by the edematous interstitial tissue.

Stomach.—The mucosa was intensely congested.

Small Intestine.—The punched out areas corresponded to parasitic infestation.

Spleen.—Focal necrosis of the lymphoid follicles was associated with a necrotizing arteriolitis.

Sections of all other organs including the placenta were normal.

Comment.—This pregnant animal injected with antiplacenta serum demonstrates the fulminating course of acute glomerulonephritis. The lesions were the most severe encountered. The necrotizing arteriolitis of the spleen was unique in the series.

N2.—Male beagle, 5 months old, weighing 9.5 kilos when injected intravenously with a total of 3 ml. per kilo of rabbit antidog-placenta serum July 8, 9, and 10, 1952.

July 11 to Aug. 7, 1952.—Proteinuria 0.2 to 1.9 gm. per cent with casts and red cells noted in the urinary sediment. Blood urea nitrogen retention (maximum 49 mg. per cent on July 24), elevation of blood cholesterol from a value of 150 mg. per cent to 250 mg. per cent, a drop in red blood cell count from 5.5 million cells per c. mm. to 4.1 million cells, and an increase in the erythrocyte sedimentation rate (maximum 37 mm./hour on July 29) occurred. A drop in total serum protein with a reversal of the albumin globulin ratio was found on 3 consecutive weekly tests. Hypertension and edema were not seen. At the end of the 2nd week of observation the dog became listless and lost 1.5 kilos in weight. On July 17 and July 22 both rabbit protein and antibody to rabbit protein were demonstrable in the blood. Serum from subsequent bleedings contained only antibody to the rabbit protein. On Aug. 7th a kidney biopsy was performed.

Aug. 8 to Day of Sacrifice, Jan. 21, 1953.—The only abnormal findings were proteinuria 0.2 to 1.7 gm. per cent, with occasional casts and red cells in the urinary sediment, and a transitory rise in blood urea nitrogen (maximum 43 mg. per cent on Sept. 11).

Biopsy, Aug. 7, 1952.—Fibrin and hyaline thrombi were present in the glomeruli. The basement membranes were irregularly thickened. An occasional glomerulus was fibrotic;
others showed adhesions between tufts and to the capsule of Bowman. Red blood cells and fibrin were present in the glomerular spaces. Lymphocytes, histiocytes, and plasma cells were seen throughout the interstitial tissue but especially about the glomeruli. The capsular as well as the glomerular epithelium was swollen and showed cellular proliferation. No arterial or arteriolar lesions were seen. The tubules were dilated and the lining epithelium was correspondingly flattened. Hyaline and very occasional red blood cell casts were present.

**Autopsy, Jan. 21, 1953.**—The beagle was an adult male in good condition weighing 9 kilos. The kidneys appeared pale with a few fine scars over the surface. The left weighed 35.4 gm.; the right, 32 gm. All other organs appeared to be essentially normal.

**Microscopic.**—

*Kidneys.* The biopsy site was well healed. There was a diffuse glomerular lesion. Many glomeruli were fibrotic and others were hyalinized. Most of the basement membranes were thickened by a hyaline deposit which obliterated many of the capillary loops. Bowman's capsule was also thickened by fibrous tissue and hyaline deposits and was often adherent to the glomerular tufts. This resulted in fine cortical scarring which was enhanced by the tubular atrophy and distortion. Comparison of the sections of the kidney obtained at autopsy with those from the biopsy showed definite progression of the glomerular lesion in that the fibrous tissue became more abundant and denser. The basement membranes in general were thicker. Dilated convoluted tubules with casts seen in biopsy were largely replaced by normal tubules. Casts were much less prominent. The cellular infiltration was similar.

**Comment.**—This male beagle injected with antiplacenta serum and subjected to biopsy 1 month later was sacrificed 6 months after injection. Histologic study illustrates definite progression of the kidney lesion to the chronic phase although the animal had no nitrogen retention at the time of death.

**A14.**—Female beagle, 7 weeks pregnant, 17 months of age, weighing 11.0 kilos when injected intravenously with a total of 3 ml. per kilo of rabbit antidoog-placenta serum Jan. 29, 30, and 31, 1952.

**Feb. 1 to 28, 1952.**—Proteinuria 0.5 to 1.4 gm. per cent, casts and red blood cells in urinary sediment, and urea nitrogen retention (maximum 114 mg. per cent on Feb. 8) were observed. Anemia, an elevated erythrocyte sedimentation rate, and an elevated serum cholesterol were present, which may have been due to the pregnancy. On Feb. 6 the total serum protein was 6.2 gm. per cent, the albumin 3.0, and the globulin 3.2 gm. per cent. Hypertension and edema were not found. Listlessness, weakness, and the loss of 1 kilo in weight, associated with vomiting led to subcutaneous administration of Cutler's solution on Feb. 8 and 9. The dog delivered 3 dead and 2 viable pups on Feb. 19th. The latter survived only 1 week. The mother had 2 infected mammary glands and was treated successfully with terramycin.

**Mar. 1 to Day of Death, Dec. 6.**—Proteinuria, 0.036 to 0.5 gm. per cent, with casts and red cells in the urinary sediment was present. Other laboratory findings including blood urea nitrogen were normal except for the expected rise in blood cholesterol (321 mg. per cent) and erythrocyte sedimentation rate (90 mm. in 1 hour) with a drop in red cell count (4.57 million cells per c. mm.) following estrus in June. The animal was killed in a canine battle in December.

**Autopsy, Dec. 7, 1952.**—There were deep tears in both groins exposing the muscles of the thigh and abdomen and smaller lacerations of the axillae. The thoracic muscles were hemorrhagic. Each kidney weighed 27 gm. The surface was finely pitted and the corticomedullary line appeared wide. All other organs appeared normal.

**Microscopic.**—

*Kidneys.*—There was a mild glomerular lesion affecting almost every glomerulus, charac-
terized by irregular thickening of the basement membrane and adhesion to the capsule. Often the glomerular capillaries were distorted by hyaline-like deposits which were positive in periodic acid-Schiff and Oil Red O preparations. Tubular damage and interstitial scarring were slight and only a few hyaline casts and some granular precipitate were present in the lumens. The blood vessels were normal. Except for postmortem autolysis all other organs were normal.

Comment.—The protocol of this animal illustrates the persistence (10 months) of symptoms and lesions of a mild nephritis, following a severe acute nephritis due to antidor-g-placenta serum.

A26.—Female beagle, 17 months of age, weighing 9.3 kilos when injected intravenously with a total of 3 ml. per kilo of rabbit antidor-kidney serum, Aug. 13, 14, and 20, 1951. Third injection of serum delayed 6 days because of transitory anemia (Hb 11.8 gm., red blood cells 4.88 million).

Aug. 14 to 24.—Proteinuria not in excess of 0.036 gm. per cent with rare casts in urinary sediment and moderate hypertension (maximum 166/104 mm. Hg). From Aug. 21 there was anemia (lowest value Aug. 24th 10 gm. per cent), and simultaneously an increased erythrocyte sedimentation rate (maximum 90 mm. in 1 hour). Blood urea nitrogen was normal.

Aug. 25 to Day of Sacrifice, Aug. 30.—Marked proteinuria (maximum 5.7 mg. per cent) associated with all types of casts, weight loss of 1.3 kilos within 4 days, and a rise in blood urea nitrogen to 62 mg. per cent on Aug. 29 and 123 mg. per cent on Aug. 30. Coincidentally signs of serum disease developed, with edema of dependent parts, purpura, and stiffness of the hind legs. Precipitin tests of the serum from serial bleedings indicated that rabbit serum was diminishing and antibody to rabbit serum increasing in the circulation.

Autopsy, Aug. 15, 1951.—The body was that of an adult female beagle weighing 8 kilos. The mucous membranes were pale. There was no free fluid in the body cavities. The heart weighed 68 gm. and was not hypertrophied. There were several mucosal erosions in the stomach with hemorrhagic bases. The kidneys weighed 34 and 36 gm. and appeared swollen. The surfaces were studded with pinpoint hemorrhages which extended throughout the cortex. The cortical striations were indistinct. All other organs appeared normal.

Microscopic.—

Kidneys.—Almost every glomerulus was damaged. Fibrinoid necrosis of tufts, adhesions to the capsule, hemorrhage in the capsular space were all present. Proliferation of glomerular and capsular epithelium as well as occasional mitosis of epithelial cells was seen. The basement membranes of glomeruli and capsules were thickened by hyaline deposits. There was an infiltration of polymorphonuclear cells, lymphocytes, and plasma cells within and about the glomeruli. There was a necrotizing lesion of a single arteriole of the kidney. The tubules were filled with hyaline and red blood cell casts.

Stomach.—The bases of the ulcers were covered with fibrin. Throughout there was an extensive necrotizing arteritis even in submucosal arteries beneath the intact mucosa. This was characterized by a fibrinoid necrosis of the vessel wall and an extensive infiltration of polymorphonuclear cells, lymphocytes, and eosinophiles. The remainder of the gastrointestinal tract showed no vascular lesions.

Synovia.—There appeared to be a slight increase in lymphocytes and histiocytes but no vascular lesions were present.

Liver.—Extrahepatic hematopoiesis and small clumps of hemosiderin pigment in wandering cells were seen.

Bone Marrow.—Appeared hyperplastic.
Comment.—This animal was sacrificed in uremia on the 10th day following the last injection of antikidney serum. The nephritis was exacerbated by serum sickness. Despite the fact that the animal was sacrificed at the height of the serum sickness the only arterial lesions were a necrotizing arteritis of the stomach and arteriolitis of a single vessel in one kidney. The kidney lesion was that of a severe acute glomerulonephritis.

N23.—Male beagle, 8 months old, weighing 11.6 kilos when injected intravenously with a total of 3.5 ml. per kilo of rabbit antidog-kidney serum, Feb. 9, 10, and 11, 1953.

Feb. 12 to March 10.—Proteinuria, 0.5 to 2.3 gm. per cent, with casts and red cells in the urinary sediment. Urea nitrogen retention was noted first on Feb. 19th (maximum 72 mg. per cent on Feb. 24). Progressive weight loss of up to 1.3 kilos, anemia (lowest red blood cell count 4.00 million cells per c. mm. on March 2), transitory rise of erythrocyte sedimentation rate (maximum 123 mm. in 1 hour on Feb. 24), and elevation of total blood cholesterol, from baseline level of 174 to 267 mg. per cent were observed. March 10th, biopsy of kidney.

Mar. 11 to Day of Sacrifice, May 13.—Proteinuria, 0.6 to 1.7 gm. per cent, with all types of casts and red cells in the urinary sediment was seen. The blood urea nitrogen continued to rise and was at a maximum (176 mg. per cent) the day the animal was sacrificed. Total cholesterol increased (maximum 352 mg. per cent on May 13) and blood pressure determinations on May 12 and 13 showed moderate hypertension (146/96 and 158/100 mm./Hg). Coincidentally the weight dropped an additional 2 kilos. The dog appeared to be dying and was sacrificed on May 13.

Biopsy, Mar. 10, 1952.—There was severe damage to the glomeruli and tubules. Some glomeruli were obliterated by fibrous tissue; others were adherent to the capsule. Basement membranes were thickened. There was proliferation of epithelial cells of both capsule and tufts. The patent capillaries appeared dilated. The tubular damage was commensurate with that of the glomeruli. Many tubules were dilated; others were atrophic. The epithelium of the convoluted tubules were swollen. There were hyaline and cellular casts. Periglomerular and interstitial infiltrations of lymphocytes and wandering cells were present. No vascular lesions were seen.

Autopsy. Sacrificed in Uremia. May 13, 1952.—Weight 8 kilos. The body was emaciated. The mucous membranes were pale. There were several shallow buccal ulcers covered with yellow slough. There was no free fluid in the chest or abdomen. The lungs were studded with petechial hemorrhages up to 4 mm. in diameter. There was no pulmonary edema. The heart weighed 85 gm. and was not hypertrophied. There was no excessive pericardial fluid and no exudate over the pericardium. A small erosion was noted in the stomach. All other organs appeared normal.

The kidneys were symmetrically enlarged. The left weighed 34.2 gm.; the right 33.5 gm. The parenchyma bulged above the cut capsule. The striations were indistinct. The pyramids were sharp. The uterus and pelvis were normal.

Microscopic.—The salient features were the following:

Kidney.—The renal lesions had progressed when compared to the biopsy sections although no acute glomerular lesions were present. The thickening of the basement membranes of capsules and glomeruli and the adhesions between the two had increased. Many glomeruli had become fibrous or hyaline balls. Tubular atrophy and dilatation had become more severe. Calcium deposits in the epithelium and lumens of the tubules as well as casts were seen. An occasional afferent arteriole was thickened by hyaline deposits or was necrotic. (The animal had a moderate hypertension terminally.)
Glomerulonephritis produced by specific antisera. II

Buccal Mucosa.—Surface epithelium was ulcerated. The base was covered with necrotic debris over granulation tissue which was heavily infiltrated with polymorphonuclear cells.

Stomach.—A phlegmonous edema of the submucosa and mucosa accompanied by a fibrinoid necrosis of the small arteries was present.

Comment.—This dog illustrates progression of acute nephritis to a chronic form terminating in uremia 90 days following the injection of rabbit antidor-kidney serum. The renal lesion appeared to be more severe in the autopsy material 2 months after biopsy. The fibrinoid necrosis of the small arteries and arterioles in the stomach and kidneys bears a resemblance to that seen in the malignant phase of human hypertension.

41.—Female beagle, age 33 months, 3 weeks pregnant, weighing 13.4 kilos when injected intravenously with a total of 4.0 ml. per kilo of normal rabbit serum Mar. 22, 23, and 24, 1951.

Mar. 25 to Apr. 2.—There were no abnormal findings.

Apr. 3 to Apr. 10.—Serum sickness, characterized by swollen, stiff, painful joints, edema, conjunctivitis, purpura, and enlarged lymph glands. Proteinuria (maximum 1.26 gm. per cent on Apr. 9) with occasional casts and red cells in the urinary sediment, anemia (lowest red cell count 3.91 million cells per c. mm. on Apr. 6), and a rise in erythrocyte sedimentation rate (maximum 107 mm. in 1 hour on Apr. 4) also occurred. Neither hypertension nor urea nitrogen retention was noted. Precipitin tests carried out on serum obtained during this period showed that the rabbit serum was decreasing while antibody titer to rabbit serum was rising in the circulation.

Apr. 11 to Day of Sacrifice, Aug. 2, 1951.—Anemia and elevation in the erythrocyte sedimentation rate persisted through the remaining 4 weeks of gestation. On May 6, 5 healthy pups were delivered which grew to maturity. Proteinuria 0.036 to 0.34 gm. per cent with occasional red cells in the urinary sediment was present throughout the 4 months of observation.

Autopsy, Aug. 2, 1951.—The body was that of a well developed and nourished female beagle weighing 14.1 kilos. No abnormalities were noted in the gross.

Microscopic.—

Kidney.—There was a healed infarct in the right kidney. The renal arteries in the section appeared normal. However, in another section one renal artery showed fragmentation of the elastica. The glomeruli showed a generalized diffuse thickening of the basement membrane. Some were atrophic and a pale coagulum was present in the capsular spaces. No arteriolar lesions were seen.

Aorta.—A single small branch of the aorta showed proliferation of the intima and fragmentation of the elastica in the media beneath. There were fragmentation and irregularity of the external elastic lamella of one of the vasa vasorum of the aorta. Synovia from several joints of the extremities were entirely normal.

Comment.—This animal, injected with normal rabbit serum, had a typical attack of serum sickness and developed proteinuria at that time which continued during the 4 month period of observation. The foreign serum may have caused the arteritis which had largely healed at the time of autopsy. There was a generalized thickening of the glomerular basement membranes which may also be consistent with a reaction to foreign serum.
M. BEVANS, B. C. SEEGAL, AND R. KAPLAN

BIBLIOGRAPHY

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EXPLANATION OF PLATES

PLATE 93

Fig. 1. Dog A39. Died in uremia 11 days after injection with antiplacenta serum. Acute necrosis of a glomerulus with destruction of the basement membrane. Periglomerular and intraglomerular cellular infiltration consists of lymphocytes, plasma cells, polymorphonuclear cells and red blood cells. Fibrin thrombi occlude some of the glomerular capillaries. There is necrosis of the arteriole entering the glomerulus. Hematoxylin and eosin stain. X 270.
(Bevans et al.: Glomerulonephritis produced by specific antisera. II)
Fig. 2. Dog M7. Acute glomerulonephritis 9 days after injection with antiplacenta serum. The glomerular basement membranes are swollen. Polymorphonuclear leukocytes, lymphocytes, and plasma cells are present in the tufts and in the interstitial tissue. The tubular epithelium is swollen and, in some areas, desquamated. Casts are seen in the lumens of dilated tubules. Necrosis of the wall of a small artery and cellular reaction appear at the left. Similar lesions in the arterioles are indicated by the black streaks near the glomeruli at the extreme right. The arteriole of the glomerulus at the upper right shows only focal areas of necrosis. Hematoxylin and eosin stain. × 300.
(Bevans et al.: Glomerulonephritis produced by specific antiserum, II)
FIG. 3. Dog A26. Acute glomerular nephritis and arteriolitis in a dog 10 days after injection with antikidney serum. The nephritis was exacerbated by the presence of manifest serum sickness. The arteriole at the bottom shows fibrinoid material and cellular infiltration about it. Two giant cells are seen in the glomerulus at the right. Hematoxylin and eosin stain. X 270.
(Bevans et al.: Glomerulonephritis produced by specific antisera. II)
FIG. 4. Dog N23. Kidney biopsy taken 30 days after injection of antikidney serum. Note the capsular proliferation and adhesion between capsules and tufts. Casts in the lumens of the tubules and hyaline droplet degeneration of the tubular epithelium are apparent. Lymphocytes and histiocytes infiltrate the interstitial tissue. Hematoxylin and eosin stain. × 150.
(Bevans et al.: Glomerulonephritis produced by specific antisera. II)
FIG. 5. Dog N23. Kidney section taken at autopsy for comparison with Fig. 4.
Died in uremia 90 days after injection with antikidney serum. The glomerular damage has increased; one is obliterated. Fibrous thickening of the capsule and dense adhesions of the tufts and capsules are seen. Atrophic tubules and casts are numerous. Cellular infiltration and scarring of the interstitial tissue are not appreciably increased. Hematoxylin and eosin stain. × 150.
(Bevans et al.: Glomerulonephritis produced by specific antisera. II)
PLATE 98

Fig. 6. Dog N2. Kidney biopsy taken 30 days after injection with antiplacenta serum. There is a glomerular lesion of moderate severity as well as swelling of the tubular epithelium. Dilated tubules with casts are seen at the right. Hematoxylin and eosin stain. × 150.
(Bevans et al.: Glomerulonephritis produced by specific antisera. II)
PLATE 99

Fig. 7. Dog N2. Kidney section taken at autopsy 180 days after injection showing persistence of the glomerular lesion. Compare with Fig. 6. Although some of the tubules are dilated most of the epithelium appears to be normal. A cast is present in one. Hematoxylin and eosin stain. × 150.
(Bevans et al.: Glomerulonephritis produced by specific antisera. II)
FIG. 8. Dog A14. Killed in a dog fight 315 days after injection with antiplacenta serum. There is a mild glomerular lesion with generalized thickening of the basement membrane and adhesions of the tufts to Bowman’s capsule. Hematoxylin and eosin stain. × 150.

Fig. 9. Dog A26. Necrotizing arteritis of the stomach. Uremia 10 days after injection with antikidney serum. The wall is completely disrupted and the lumen occluded by a thrombus. The cellular infiltration includes polymorphonuclear neutrophiles, eosinophiles, and lymphocytes. Hematoxylin and eosin stain. × 150.
(Bevans et al.: Glomerulonephritis produced by specific antisera. II)
PLATE 101

Fig. 10. Dog 41. Injected with normal rabbit serum. Killed under anesthesia at 42 days. The basement membranes of the glomeruli are thickened. Periodic acid-Schiff stain. × 270.

Fig. 11. Dog 41. Vas vasorum in adventitia of aorta. Note slight distortion particularly of external elastic lamella. This was the only dog of the control group to develop frank serum sickness. Elastic Van Gieson stain. × 450.
Bevans et al.: Glomerulonephritis produced by specific antisera. II