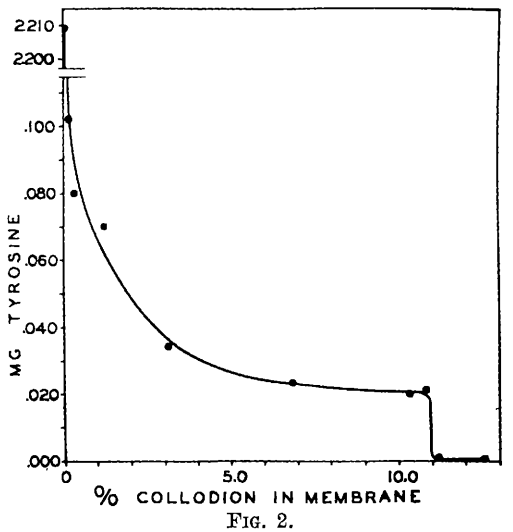

FIG. 2.

Decrease in trypsin activity as the concentration of collodion in the membranes is increased. Trypsin activity is expressed as mg of tyrosine.

the membranes is shown by the controls.

The high degree of sensitivity of the controls is illustrated in Table I from the differences between the same kind of intact and defective membranes. Thus, in the case of the cellophane membranes, the intact membrane transferred no tryptic activity, nor did the controls indicate leaking. However, the defective membrane showed a tryptic activity value of .020, while its controls jumped to .008 and .006 indicating leakage. Similar results were obtained for intact and defective 0.19, 10.4 and 11.2% collodion membranes. Therefore, experiments yielding no leak values but yielding transfer values from .020 all the way to .160 must require the interpretation that tryptic activity had been transferred across the membranes in some manner. In Rothen's experiments the thickness of the films cast on smooth surfaces may be measured with considerable accuracy by the ellipsometer. The thickness of the films in the interstices of the impregnated membranes is purely a matter of conjecture. However, both methods indicate transference of tryptic activity across thin membranes. Rothen's findings indicate that interaction takes place in systems wherein enzyme, substrate and barrier are immobilized as solid films. The experiments here described indicate that the effect also takes place when enzyme and substrate are in solution and are separated by a solid membrane. Whatever the mechanism for the transfer of the activity may be, the latter experiments show that this effect would have to be ascribed to some phenomenon other than simple diffusion through the membranes.

Summary and Conclusions. Formvar, parlodion and collodion membranes have been prepared by impregnating paper with solutions of these substances. A suitable cell of two compartments was constructed in which solutions of trypsin and hemoglobin could be separated by the membranes.

Interaction between solutions of trypsin and hemoglobin separated by formvar, parlodion or collodion membranes has been demonstrated.

Examination of a series of membranes impregnated with collodion of different concentrations showed that interaction decreased in a curvilinear fashion as the collodion concentration in the membranes was increased.

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Antigenic Property of Renal Cortex.* (17690)

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The experimental production of chronic renal disease in rats by means of heteronephrotoxins obtained from rabbits has been studied by Smadel, Swift and Farr(1-3), and later

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1. Smadel, J. E., and Farr, L. E., *J. Exp. Med.*, 1937, v65, 527.

TABLE I.

Nephrotoxicity of Sera Derived from Rabbits Treated with Extracts from Kidneys, Renal Cortex, and Renal Medulla Obtained from Rats.

Whole kidneys			Renal Cortex			Renal Medulla		
No. of rats used	Amt of serum producing severe disease, cc	Nephrotoxicity of serum	No. of rats used	Amt of serum producing severe disease, cc	Nephrotoxicity of serum	No. of rats used	Amt of serum producing severe disease, cc	Nephrotoxicity of serum
7	?	—	8	?	—	7	?	—
7	?	—	4	.9 -1.15	++	7	?	—
9	.85-1.2	++	12	.77-1.0	++	7	?	—
14	.77-1.0	+++	3	.5 - .7	+++	7	?	—
10	.77-1.0	+++	7	.51- .7	+++	6	?	—
15	.51- .7	+++	6	.38- .5	+++	15	?	—
5	.38- .5	+++	3	.38- .5	+++	5	1.5-2.1	(+)
7	.38- .5	+++	18	.3 - .4	+++			
12	.38- .5	+++	4	.2 - .3	++			
19	.19- .26	++						

on by Heymann and Lund(4-5). The former investigators described this disease as a diffuse chronic and progressive glomerulonephritis while Heymann and Lund believe that it simulates lipemic nephrosis, with and without glomerular lesions(6) as observed in infants and children. Regardless of these differences in the concept of nomenclature and classification of this disorder, studies were undertaken to determine whether renal cortex and renal medulla differ in their antigenic properties.

Methods. Renal extracts were prepared from perfused rat kidneys according to the technic previously described(4,5). The separation of cortex and medulla was approximate and was done by sharp dissection. Rabbits were injected with 10 cc of these extracts intraperitoneally every other day. 10%, 15% and 20% extracts were used for the first 3 treatments, and 20% suspensions were used for 2 more weeks. The animals were bled 6 - 7 days after the last extract injection. Rats of the Long-Evans and Whelan strains were used. They were kept on Friskies. They were injected intravenously, when 4 - 5 weeks old and weighing 50 - 70 g, with 1/3rd of the dose on 3 consecutive days. The serum had been inactivated for ½ an hour at 56°C. The amount of serum was gauged according to the estimated kidney weight as previously described(5). The dose was then varied until the amount had been established that produced a "severe" nephrotic disease. A severe renal disease, the term used in Table I, was assumed when the majority of the injected rats showed ascites and edema at onset and massive 4+ albuminuria which did not subside for 8 weeks.

Results. From 26 rabbits used, 10 were treated with extracts prepared from whole kidneys, 9 with extracts from renal cortex and 7 were injected with extracts prepared

2. Smadel, J. E., *J. Exp. Med.*, 1937, v65, 541.

3. Smadel, J. E., and Swift, T. F., *J. Exp. Med.*, 1941, v74, 345.

4. Heymann, W., and Lund, H., *Science*, 1948, v108, 448.

5. Heymann, W., Lund, H. F., Gilkey, C., and Salehar, M., Lipemic Nephrosis in Rats.

6. Heymann, W., and Alperin, L. J., *Am. Practitioner*, 1949, v3, 650.

from renal medulla obtained from rats. The sera of these rabbits were injected intravenously in 224 rats. It can be seen (Table I) that 8 of 10 sera obtained from rabbits treated with kidney extracts were nephrotoxic. The amount of serum needed to induce severe renal disease was from 0.19 to 1.2 cc. 8 of the 9 sera obtained from rabbits treated with cortex extracts were nephrotoxic. The dose needed to induce severe renal disease was from 0.2 to 1.15 cc. From 7 sera obtained from rabbits treated with medulla extracts only one was slightly nephrotoxic. The amount of serum needed to induce severe renal disease with that serum was greater than any of the least nephrotoxic sera of the kidney or cortex group. The remaining 6 medulla sera were tested in doses of 2 to 3 cc and no proteinuria was noted in any of the rats. The clinical course was carefully studied in all animals. The systolic blood pressure was taken every 2-3 weeks. B.U.N. determinations were carried out repeatedly intra vitam, and histological examinations of the kidneys and blood chemical studies were obtained at time of death, which included B.U.N., creatinine, serum proteins, cholesterol and total lipids. No differences were noted whether renal disease had been induced by using sera obtained from rabbits treated with whole kidney, cortex or medulla extracts. Two additional rabbits were treated with medulla extracts that were diluted in a ratio of 2 to one with 20% extracts of perfused rat spleens. The sera of these two animals did not produce proteinuria in 8 rats.

Discussion. The results indicate that the antigen of renal tissue is localized in the renal cortex. The nephrotoxicity of serum obtained from one of seven rabbits treated with medulla extracts may well be due to the fact that the separation of cortex and medulla was only approximate. Medulla extracts might at times have contained some cortex tissue. These results are in accordance with those obtained by Pressman, Hill and Foote(7), who studied radio autographs of kidneys after the injection of radio-iodinated globulin of anti-mouse-kidney sera. They showed localization of I^{131} in the glomeruli. The observations also are consistent with results reported lately by Solomon, Gardella, Panger, Dethier and Ferrehee(8) that nephrotoxins against rat kidneys can be removed by adsorption with suspensions of rat glomeruli, but not by adsorption with other portions of rat kidney. The specificity of this phenomenon remains, however, questionable as the adsorption of other than renal antibodies was not included in that study.

The histology of glomerular tissue shows an intimate mixture of tufts of capillary loops with epithelial elements probably deriving from structures of the proximal convoluted tubules. As the sera of rabbits treated with a mixture of medulla and spleen extracts were not nephrotoxic, and considering the highly differentiated functional state of the glomerular capsule and the proximal convoluted tubules, the latter structures might be more likely to harbor the antigen than the tufts of capillary loops.

Summary. 1. Ten rabbits were treated with extracts of bloodfree rat kidneys. Eight of the sera obtained from these rabbits induced severe renal disease in rats, weighing 50-70 g, when injected in a dose of 0.19 to 1.2 cc.

2. Nine rabbits were treated with extracts of renal cortex obtained from rats. Eight of the sera obtained from these rabbits induced severe renal disease in rats, weighing 50-70 g, when injected in a dose of 0.2-1.15 cc.

3. Seven rabbits were treated with extracts of renal medulla obtained from rats. Only one of the sera obtained from these rabbits induced severe renal disease in rats weighing from 50-70 g when injected in a dose of 1.5-2.1 cc.

4. It is concluded that the antigen of renal tissue is localised in the renal cortex.

7. Pressman, D., Hill, R. F., and Foote, F. W., *Science*, 1949, v109, 65.

8. Solomon, D. H., Gardella, J. W., Paugh, H., Dethier, F. M., and Ferrehee, J. W., *J. Exp. Med.*, 1949, v90, 267.

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