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The Zone of Localization of Anti-Mouse-Kidney Serum as Determined by Radioautographs[1]

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It has been shown previously by Pressman and Keighley (*4*) that antiserum prepared against rat kidney actually localizes in the kidney after intravenous injection into a rat. This was accomplished by iodinating the globulin fraction of anti-rat-kidney serum, prepared according to the method of Smadel (*5*), with iodine containing tracer amounts of I^{131} and following the localization of the radioactivity in the kidney of the rat inoculated with this preparation. Similar results with mice have since been obtained with antiserum prepared against mouse-kidney tissue (*3*). By using iodine with a sufficient admixture of I^{131}, it has been possible for us to obtain radioautographs of the kidney tissue of mice injected intravenously with the radioiodinated globulin fraction of anti-mouse-kidney serum, and thus to determine more precisely the region of localization within the kidney.

In the experiment reported here, two mice were injected intravenously with the radioiodinated globulin fraction of anti-mouse-kidney serum prepared similarly to the radioiodinated globulin fraction of anti-rat-kidney serum described previously (*4*). Two control mice were injected with the radioiodinated globulin fraction of anti-mouse-plasma serum, prepared similarly, using the serum from rabbits which had been injected with mouse plasma. Such serum does not localize in the kidney as does the anti-mouse-kidney serum. Each mouse injected with the antikidney preparation received 0.3 ml of solution containing 3.3 mg of protein combined with 17 μc of radioiodine and, in the case of the mice injected with the antiplasma fraction, 4.8 mg of protein, combined with 13 μc of radioiodine in 0.3 ml of solution, was used. Five days after the injection the animals were sacrificed. A blood sample was obtained before death from the dorsal aorta, and then the kidneys, spleen, and liver were removed without perfusion. One kidney, one half of the liver and spleen, and all the blood were used in the preparation of samples for the determination of the radioactivity content of the tissue. The results are given in Table 1. The other kidney and the rest of the spleen and the liver were fixed in 10% formalin and subsequently sectioned. Sections 10-μ thick and the blocks remaining after several sections had been cut were used in the preparation of radioautographs. The blocks were set up on Eastman Kodak medium lantern slides, while the sections were set up on Ansco No Screen X-ray film, according to the method described by Marinelli and Hill (*2*).

TABLE 1

RADIOACTIVITY OF TISSUES OF MICE INOCULATED WITH RADIOANTISERA 5 DAYS AFTER INOCULATION

	Antiserum used	
	Antikidney serum	Antiplasma serum
Protein inoculated (mg)	3.3	4.8
Radioiodine on protein at the time of injection (μc)	17	13
Tissue	**Activity of tissue (μc/gram, 5 days after inoculation)**	
Kidney	0.25	0.08
Liver	.08	.06
Spleen	.16	.13
Blood	.27	.21

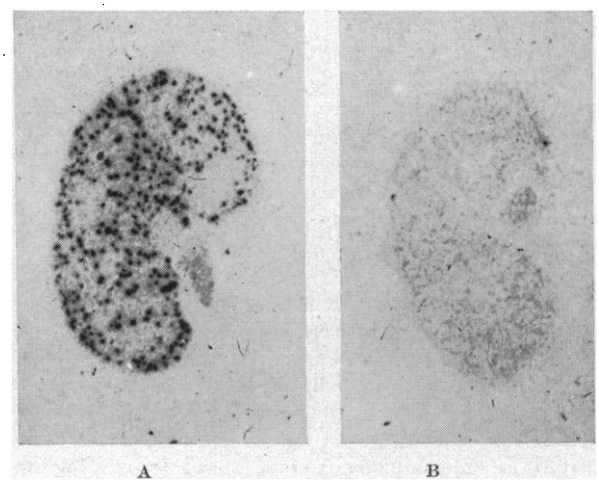

FIG. 1. Radioautographs of blocks of kidney tissue. A—from mouse injected with radioanti-mouse-kidney serum; B—from mouse injected with radioanti-mouse-plasma serum.

Fig. 1 shows the radioautographs of the kidney tissue blocks. The kidney from the mouse receiving the radioantikidney serum showed a definite accumulation of radioactivity around the cortex[3] (Fig. 1A) while the kid-

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[3] This is in accordance with the results of Heymann and Lund (*Science,* 1948, **108**, 448), who found that nephrotoxic antisera are produced from the cortical rather than the medullary kidney tissue.

ney from the mouse receiving the radioantiplasma serum showed no such accumulation (Fig. 1B). No accumulation was observed with either the liver or the spleen of the animals receiving the antikidney serum or the antiplasma serum. Fig. 2 shows the radioautographs of the kidney sections. In the sections from the animal receiving the radioantikidney serum there was a punctate accumulation of the radioactivity (Fig. 2A), while this was not the case for the kidney of the mouse receiving the radioantiplasma serum (Fig. 2B). Also, there was no such accumulation of radioactivity in the spleens or livers of the animals receiving either radioantikidney or radioantiplasma serum. With these tissues, very faint and diffuse radioautographs were obtained due to the radioactivity content of the blood in the organ.

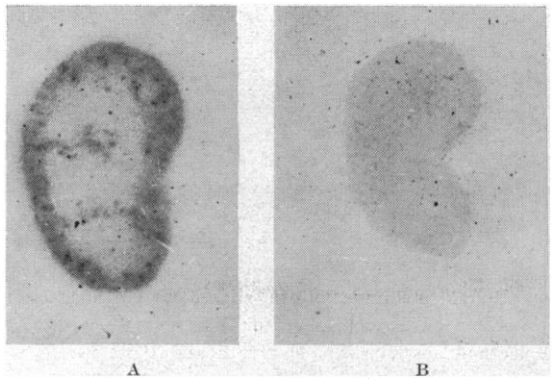

FIG. 2. Radioautographs of sections of kidney tissue. A—from mouse injected with radioanti-mouse-kidney serum; B—from mouse injected with radioanti-mouse-plasma serum.

Upon comparing the microphotograph of the kidney from the animals receiving the radioantikidney serum with an enlargement of the radioautograph, it was quite clear that the localization of the radioactivity and presumably the radioantibody was taking place in the glomeruli of the kidney. This point will be discussed more fully elsewhere (*3*).

The concentration of the antikidney serum in the glomeruli is not due to a nonspecific pickup by the kidney of foreign substances or to the fact that the radioactive material may pass through the kidney in its excretory path. The evidence for this is the fact that the radioantiplasma serum control contains substances of the same nature as radioantikidney serum, except for the specific kidney antibodies, and does not show any localization in the kidney. Similarly negative results were shown by a radioantiovalbumin serum. The radioactivity concentration in the various tissues for the two sera described here were quite similar except in the case of the kidney (Table 1). There the concentration of the radioactivity in the kidney of the animal receiving the radioantikidney serum was about three times that in the kidney of the animal receiving the control serum. The radioactivity in the kidney of the mouse receiving the control serum was essentially all due to the blood in the kidney, since mouse kidneys contain about 30–35% blood (*1*). The greater amount of radioactivity in the kidneys of the animal which received the antikidney serum must have been due to the kidney antibodies.

The localization of I^{131} in the glomeruli as shown by the radioautographs is probably a result of the corresponding localization of the specific antibodies to glomerular tissue.

These experiments were repeated with other preparations of radioantikidney serum with similar results.

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Growth of Potato Sprouts Retarded by 2,4,5-Trichlorophenoxyacetic Acid[1]

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Guthrie (*1*) first discovered that methyl ester of α-naphthalene acetic acid, when applied to potato tubers, retards the growth of sprouts. Many potato growers are now using this chemical on a commercial scale to prevent sprouting of potatoes in storage. Smith, Baeza, and Ellison (*2*) found during the 1945 season that this chemical also retards sprout growth of potatoes in subsequent storage when it is applied as a spray to the potato plants during the growing season. During the 1946 season the authors found that two spray applications of 2,4,5-trichlorophenoxyacetic acid (2,4,5-T), 25 ppm on July 19 and 50 ppm on August 20, retarded subsequent sprout growth of potatoes to the same degree as two applications of methyl ester of naphthalene acetic acid (MENA), 2,000 ppm on July 19 and 2,000 ppm on August 20.

In the spring of 1947 tubers of the Sebago variety were treated separately with MENA and 2,4,5-T in isopropyl alcohol and water and applied as a spray at the rate of 1 gm of chemical/bushel of potatoes. Although both chemicals significantly retarded sprout growth as compared with untreated tubers, sprouts of those treated with MENA had significantly less weight than those of tubers treated with 2,4,5-T.

During the 1947 growing season spray applications of sodium naphthalene acetate (sodium NA), 500, 1,750, and 3,000 ppm, compared with applications of sodium 2,4,5-T at 50, 175, and 300 ppm, were made to plants of the Sebago variety. The effect on sprout growth during subsequent storage at 50° F from October 25 to February 26 is shown in Table 1.

In all cases sodium 2,4,5-T retarded sprout growth to a greater degree than sodium NA, although the latter was applied in concentrations 10 times the former. There

[1] Paper No. 301, Department of Vegetable Crops, Cornell University.