TABLE I. Effect of Aureomycin Given by the Intravenous Route on Coagulation Time.

Patient No.	Clotting time (min.) Before aureomycin	After aureomycin	Diff.
1*	30	19.5	—10.5
2	21	15	— 6
3	19	17	— 2
4	19	17.5	— 1.5
5	17	13	— 4
6	22	15.5	— 6.5
7	23	19.5	— 3.5
8	23	21	— 2
9	26	23	— 3
10	23	23	0
11	16	12	— 4

* Convalescing from infectious hepatitis.

The data presented is in accord with Macht and Farkas' finding(1) that in certain instances aureomycin increases the coagulability of the blood. The present results suggest that this increased coagulability is due to an anti-heparin effect of aureomycin. The observation of Miller *et al.* that certain antibiotics including aureomycin reduce mortality from x-radiation(6) and the finding of Allen *et al.* that shortly after x-radiation a heparin-like substance appears in the plasma of dogs (7), suggest the possibility that protection by certain antibiotics against the effects of radiation may be mediated, at least in part, through an anti-heparin effect. Whether or not this phenomenon is responsible for an increased incidence of thrombosis following the use of antibiotics, which has been suggested by some observers and denied by others(8), should be investigated further. It is worthy of note that both streptomycin and penicillin have been shown to increase the coagulability of blood(9-11).

Summary. Addition of aureomycin to human plasma *in vitro* produces a significant decrease in the heparin concentration. Aureomycin administered by the intravenous route lowered the coagulation time of the blood of 10 of 11 patients.

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6. Miller, C. P., and Hammond, C. W., *Science,* 1950, v111, 719.

7. Allen, J. G., Sanderson, M., Milham, M., Kirschon, A., Jacobson, L. O., *J. Exp. Med.,* 1948, v87, 71.

8. Long, P. H., *J. Am. Med. Assn.,* 1950, v142, 49.

9. Moldavsky, L. F., Hasselbrock, W. B., Cateno, C., *Science,* 1945, v102, 38.

10. Macht, D. I., *Science,* 1947, v105, 313.

11. Macht, D. I., *Arch. Int. of Pharmacodyn.,* 1947, v74, 399.

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Effects of X-irradiation on Passively Transferred Antibody.* (18237)

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Although much work has been done in an attempt to elucidate the mechanisms involved in antibody production and the factors which influence active immunity, very little is known about the mechanism of antibody destruction. Heidelberger *et al.*(1) have demonstrated that dietary amino acids containing isotopic nitrogen (N 15) are incorporated into actively produced antibody in similar quantities to their incorporation into the plasma proteins, and that the rate of disappearance from the 2 proteins is similar. Passively transferred homologous antiserum showed no uptake of the dietary isotopic nitrogen and was therefore considered anabolically inert.

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1. Heidelberger, M., Treffers, H. P., Schoenheimer, R., Ratner, S., and Rittenberg, D., *J. Biol. Chem.,* 1942, v144, 555.

TABLE I. Rate of Disappearance of Passively Transferred Homologous Antibody.

	Time in days after injection of antiserum								
	1/24	1	2	3	4	6	8	10	12
Irradiated	1333	1000	640	600	600	400	160	75	60
	800	640	600	400	300	160	100	60	60
	1000	800	600	400	300	160	120	60	60
Control	1333	533	400	400	400	200	120	75	40
	1600	800	400	400	400	300	100	60	60
	1600	1000	400	600	400	200	100	60	40

TABLE II. Rate of Disappearance of Passively Transferred Heterologous Antibody.

	Time in days after inj. of antiserum					
	1/24	1	2	3	4	6
Irradiated	2132	800	400	200	160	40
	1600	800	400	160	160	80
	1600	533	266	200	160	20
Control	1600	800	400	160	40	20
	1600	800	266	160	80	20
	1600	800	200	160	80	40

X-irradiation has been shown to inhibit antibody formation(2). Once active antibody formation has started, however, irradiation does not alter the response of the animal to the antigen(2,4). It seemed desirable, therefore, to study the effect of x-ray on the rate of antibody destruction after passive transfer. In our experiments both heterologous and homologous antisera were used, since Glenny and Hopkins(3) have shown that the former is destroyed much more rapidly than the latter.

Materials and methods. Albino rabbits from 2-2.5 kg in weight were used in all experiments. The antisera were given intravenously 24 hours after irradiation, and control animals were injected at the same time. Blood samples for antibody titer were taken thereafter at intervals as shown in Tables I and II. The homologous antiserum was a rabbit anti-sheep red blood cell hemolysin with a titer of 1:5333, and 25 cc of the antiserum was injected intravenously into each animal. Hemolysin titers were determined by a standard technic, with excess complement and 2% sheep RBC's as indicator. The end point was taken as the last tube showing clear sparkling hemolysis after 1 hour in a water bath at 37°C. The heterologous antiserum was obtained from a patient recovering from brucellosis, and 7.5 cc of this antiserum with a titer of 12,500 was injected intravenously. Agglutinin titers were determined with equal parts of diluted serum and a heat killed antigen after 24 hours in a water bath at 56°C and 24 hours refrigeration at 8°C. Individual tube dilutions, rather than serial dilutions, were used in all experiments.

X-ray dosage was patterned after the method of Craddock and Lawrence(4), who reported that a dosage of 250 r was sufficient to depress antibody production in rabbits without producing changes in the peripheral blood or general nutrition of the animals. We used a total of 300 r with the following radiation factors: 250 Kv, 18 ma, 0.5 mm Cu and 1 mm Al filters, 50 cm distance, open port, and 72 r/min. dose rate. This small roentgen dosage was felt to be preferable in order to prevent, as much as possible, the protein catabolic effects of irradiation sickness. With the animal stretched on a board, the effective radiation, as checked with a fluorescent screen, did not include small portions of the extremities and of the ears.

Twenty-five days after the initial serum injection, anti-human serum titers were determined on the animals which had received the human brucella immune serum, using the same human brucella serum as the antigen. The undiluted rabbit serum was placed in precipitin tubes and overlayed with the antigen in dilutions of 1:5, 1:10, 1:100, 1:1000, 1:10,000, and 1:100,000. The last tube

2. Hektoen, L., *J. Infect. Dis.*, 1919, v17, 415.

3. Glenny, A. T., and Hopkins, B. E., *J. Hyg.*, 1922-23, v21, 142.

4. Craddock, C. G., Jr., and Lawrence, J. S., *J. Immunol.*, 1945, v60, 241.

showing reaction at the interphase after 20 minutes was taken as the end point.

Results. As can be seen in the accompanying tables, the rate of destruction of the homologous antibody (Table I) and the heterologous antibody (Table II) was markedly different and fell within the ranges described by Glenny and Hopkins(3). There was no difference between the irradiated and the control animals. The anti-human serum titers were determined primarily to check the efficacy of this irradiation method and dosage in inhibiting antibody formation. Results in the 3 irradiated rabbits were positive in antigen dilutions of 1:5, 1:10, and 1:10 respectively; the 3 control animals were all positive in 1:1000 antigen dilution.

Summary and conclusions. X-irradiation in dosage of 300 r had no effect on the rate of destruction of passively transferred homologous or heterologous antisera given intravenously to rabbits 24 hours after irradiation. This x-ray dosage was sufficient, however, to decrease, but not totally prevent, the formation of antibody to the heterologous serum proteins.

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Influence of Fasting on the Effect of Insulin on Glucose Oxidation.* (18238)

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It has generally been believed that insulin increases the immediate oxidation of glucose and this is frequently stated in text books(1). However as Soskin and Levine(2) point out in their monograph, "this is without basis in fact." There has been no good experimental evidence that insulin will increase the oxidation of glucose until recently(3) when by using carbon^{14}-labeled glucose it was found that the old assumption is correct. Insulin actually does increase the oxidation of glucose by the extra-hepatic tissues to carbon dioxide and water. This report concerns the influence of fasting on the effect of insulin on glucose oxidation by the extra-hepatic tissues.

Experimental. The general plan of the experiments and the details of the methods used have been given elsewhere(3-6). After varying periods of fasting male rabbits (average weight 2 kg after evisceration) were eviscerated(7) and given an injection of a priming dose of carbon^{14}-labeled glucose so as to make the specific activity of the body glucose the same as the glucose to be used for the constant injection. With the aid of frequent blood sugar determinations the blood sugar was maintained at a normal level (70-130 and as near 100 mg % as possible) for 8 or more hours by the constant injection of carbon^{14}-labeled glucose. Ten units of insulin† were given intravenously each hour. Collection of the expired carbon dioxide was commenced immediately after the injection of the radioactive glucose.

Results. The data in Fig. 1 were calculated

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1. Best, C. H., and Taylor, N. B., The Physiological Basis of Medical Practice. The Williams & Wilkins Co., Baltimore, 1943, p972.

2. Soskin, S., and Levine, R., Carbohydrate Metabolism. University of Chicago Press, p181.

3. Wick, A. N., Drury, D. R., Bancroft, R. W., and MacKay, E. M., *J. Biol. Chem.*, in press.

4. Wick, A. N., Drury, D. R., and MacKay, E. M., *Am. J. Physiol.*, 1950, v163, 224.

5. Drury, D. R., Wick, A. N., and MacKay, E. M., *Am. J. Physiol.*, in press.

6. Drury, D. R., Wick, A. N., Bancroft, R. W., and MacKay, E. M., *Am. J. Physiol.*, in press.

7. Drury, D. R., *Am. J. Physiol.*, 1935, v111, 289.

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