Feto-Maternal and Feto-Fetal Transfer of I-131 Gamma Globulin in the Rat.* (28827)

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Antibodies have been demonstrated to pass from mother to fetus both experimentally in animals(1) and clinically in human beings (2). The subject was reviewed by Edsall(3) and by Brambell and Hemmings(4). The corollary that antibodies pass from fetus to mother does not appear to have been investigated.

In the following experiments with rats, I-131 labelled gamma globulin was used to show that the passage of antibody occurred from mother to fetus, from fetus to mother, and from fetus to fetus through the maternal circulation.

Materials and methods. Gamma globulin was obtained from the sera of healthy Long-Evans rats by diethylamino-ethyl cellulose column chromatography using the method previously described(5), with 0.01 M pH 7.95 phosphate buffer. The gamma globulin solution was concentrated with carbowax (20 M polyethylene glycol) dialyzed to pH 7.5, and iodinated by the tri-iodide method(6). The presence of gamma globulin before iodination was confirmed by cellulose acetate paper electrophoresis. The nitrogen content of the gamma globulin fraction as shown by the micro Kjeldahl method was 132.0 μg nitrogen per ml before iodination and 72.0 μg nitrogen per ml after iodination. After iodination, the presence of I-131 gamma globulin was shown by autoradiography of cellulose acetate strips after electrophoresis of the I-131 gamma globulin. Of the total I-131, 95% was precipitable with 10% trichloracetic acid. The I-131 gamma radioactivity was determined with an automatic gamma counter, using a well-type scintillation detector coupled to a spectrometer.

In Experiment I, three Long-Evans rats, 21 days pregnant, were injected intravenously with 1 ml of I-131 labelled gamma globulin solution. Blood samples were obtained from the mother and fetuses at intervals to 24 hours. The fetal blood was obtained by cardiac puncture following removal of 3 or 4 fetuses from the uterus. The blood was then pooled and the fetuses sacrificed.

In Experiment 2, several fetuses in two 21 day pregnant Long-Evans rats were injected intraperitoneally with 0.15 ml I-131 labelled gamma globulin and blood samples obtained from the mother at intervals up to 6 hours. The mothers were sacrificed at the end of 6 hours and blood samples obtained by cardiac puncture from the injected and non-injected fetuses. In all preparations the feti were viable at time of sacrifice as judged by fetal heart rate, pulsation of placental vessels and fetal skin color(7).

Samples for determination of radioactivity included maternal and fetal sera, whole fetuses, and whole placentae. The protein bound I-131 in the sera was determined by precipitation with cold 10% trichloracetic acid, and by autoradiography of cellulose acetate strips after electrophoresis. All counts were 3 times greater than background except for some in rats A and B, which are indicated by an asterisk in Table III.

Results and discussion. Table I shows the

TABLE I. Total Fetal I-131 Following Intravenous Injection of Maternal Rat 1 with I-131 Gamma Globulin.

Fetus No.	Time after inj, hr	% of I-131 gamma globulin injected
1	6	.19
2	6	.75
3	6	.35
	Mean	.43
4	24	1.08
5	24	.71
6	24	1.38
7	24	.76
	Mean	.98

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TABLE II. I-131 Gamma Globulin in Maternal and Fetal Sera Following Intravenous Injection of Rats 1, 2 and 3.

Sample	Time after inj, hr	Rat 1		Rat 2		Rat 3
		% of I-131 gamma globulin inj/ml serum	% I-131 precipitated with T.C.A.	% of I-131 gamma globulin inj/ml serum	% I-131 precipitated with T.C.A.	% of I-131 gamma globulin inj/ml serum
Maternal serum	.017			6.63		6.83
	1	4.70		5.22		6.02
	2	4.39		5.03		5.65
	6	3.13		3.11	99.3	
	24	1.42	97.9			1.20
Fetal serum	3	.09		.08		
	6	.12		.17	52.8	
	24	.20	51.0			.30

amount of I-131 gamma globulin present in the feti expressed as a percentage of the total I-131 gamma globulin injected into the mother. The mean percentage recovered per fetus rose from 0.43% 6 hours after injection to 0.98% 12 hours after injection. Table II shows that following injection of 3 maternal rats, concentrations of I-131 gamma globulin decreased in maternal sera over a 24 hour period, and increased in fetal sera during the same period. Both tables show that as time progressed following injection of the mother, there was a decrease in I-131 gamma globulin in the mother, and an increase in I-131 gamma globulin in the feti.

T.C.A. precipitation demonstrated that 97.9% to 99.3% I-131 was bound to gamma globulin in the maternal serum. After passing into the fetal serum 51.0% to 52.8% of the I-131 remained bound to gamma globulin, indicating that dissociation had occurred during the passage from maternal to fetal circulations, and that some of the fetal I-131 represented iodide I-131 after degradation of the protein in the mother, or in the fetus, or during its transfer from maternal to fetal circulations.

Autoradiography of the cellulose acetate electrophoretic patterns obtained on the above maternal and fetal sera showed that I-131 was present in the gamma globulin fraction (Fig. 1).

In Experiment 2, six hours after injection, 23.3% of the I-131 gamma globulin injected was present in the 4 injected fetuses in rat B, and 0.09% was present in the 4 placentae, which included maternal blood. Approximately 6% of the original I-131 gamma globulin injected was present in the fetal blood withdrawn for examination. A total of 29.4% of the original I-131 gamma globulin was present in the fetuses and placentae 24 hours after injection. The 8 non-injected fetuses contained 1.01% of the original I-131 gamma globulin. Their placentae contained 0.04% and the blood approximately 0.025%,

TABLE III. I-131 Gamma Globulin in Maternal and Fetal Serum Following Injection of Feti in Rats A and B.

Sample	Time after inj, hr	Rat A		Rat B	
		% of I-131 gamma globulin inj/ml serum	% I-131 precipitated with T.C.A.	% of I-131 gamma globulin inj/ml serum	% I-131 precipitated with T.C.A.
Maternal serum	1	.004*		.005*	
	3	.014*		.027*	74.9
	6	.042*	70.8	.069	87.7
Serum from injected feti	6	8.32		2.28	96.9
Serum from non-injected feti	6	.026*		.039*	54.3

* Radioactivity counts less than 3 times background.

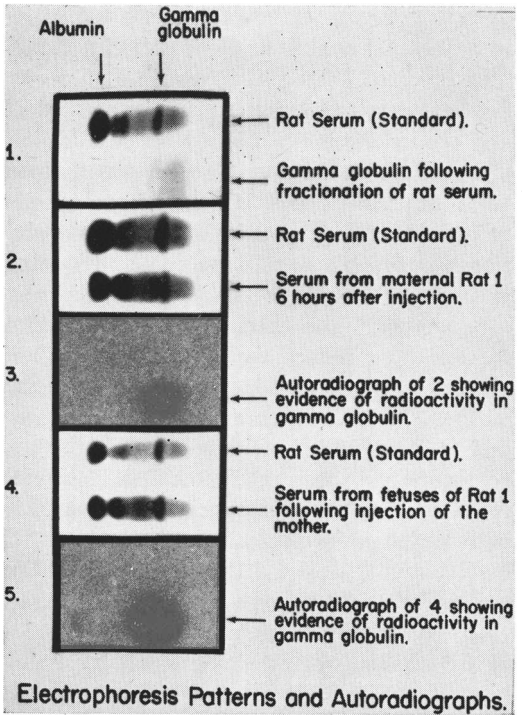

FIG. 1. Electrophoretic and autoradiographic patterns of maternal and fetal serum following administration of I-131 rat gamma globulin.

forming a total of 1.08% of the original I-131 gamma globulin recovered from these feti and placentae.

The percentage of original I-131 gamma globulin recovered from the maternal serum following intraperitoneal injection of the feti in 2 pregnant rats is shown in Table III. The percentage recovered per ml maternal serum increased during the 6 hours of the experiment in direct relation to the length of time following injection. T.C.A. precipitation showed that 96.9% of the I-131 was bound to gamma globulin in the sera of the injected feti, between 70.8 and 87.7% remained bound in the mothers' sera and 54.3% was bound in the sera of the non-injected feti. These figures indicate that the degree of dissociation of I-131 from gamma globulin increased with passage from injected feti to mother to non-injected feti.

The results indicate that in rats there is a complex interaction between the circulations of mother and fetus at the sites of closest approximation.

Summary. I-131 rat gamma globulin was shown to pass from the mother to the fetuses *in utero,* during the 24 hour period following injection of 3 maternal rats. I-131 gamma globulin was also shown to pass from the fetuses *in utero* into the maternal circulation, and thence into the circulation of the non-injected fetuses, following injection of fetuses *in utero* in 2 pregnant rats.

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Uptake of Tritiated Thymidine by Leukemic Cells: Effect of Various Leukocyte Preparations. (28828)

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Recently, Craddock has suggested that normal mature granulocytes contain a potent inhibitor of DNA synthesis which is absent from leukemic cells(1). Using dog thoracic duct lymphocytes as test cells, he demonstrated that DNA-P^{32} incorporation and thymidine-H^3 uptake were inhibited by intact or homogenized normal human granulocytes but