TABLE I. Effect of Amphetamine on Pressor Response to Tyramine.

Dose of tyramine	No. of rats	Avg pressures before tyramine (mm Hg ± S.E.) Diastolic	Mean	Systolic	Avg pressures at peak of response to tyramine (mm Hg ± S.E.) Diastolic	Mean	Systolic	Avg increase in mean pressure (mm Hg ± S.E.)
				Normal (no amphetamine)				
2.5	13	128 ± 4	143 ± 4	157 ± 4	135 ± 5	149 ± 5	165 ± 8	6 ± 4
5.0	8	119 ± 4	134 ± 3	149 ± 1	128 ± 5	148 ± 7	166 ± 10	14 ± 6
10.0	6	115 ± 6	131 ± 6	149 ± 8	139 ± 3	160 ± 6	182 ± 9	29 ± 7
25.0	6	130 ± 7	146 ± 7	163 ± 5	147 ± 4	187 ±11	215 ± 11	41 ±15
				After amphetamine, 0.5 mg/kg i.v.				
2.5	6	114 ± 7	125 ±11	138 ±11	128 ± 6	153 ± 8	174 ± 10	28 ± 3
				After amphetamine, 1.0 mg/kg i.v.				
2.5	6	119 ± 3	133 ± 4	148 ± 4	148 ± 4	168 ± 7	190 ± 8	35 ± 5
				After amphetamine, 2.0 mg/kg i.v.				
2.5	6	122 ± 5	138 ± 5	154 ± 6	138 ± 5	165 ± 7	194 ± 13	27 ± 6
				After amphetamine, 4.0 mg/kg i.v.				
2.5	6	122 ± 3	138 ± 4	155 ± 6	125 ± 6	148 ± 9	171 ± 12	10 ± 7
				After amphetamine, 2.0 mg/kg i.d.				
2.5	7	113 ± 5	128 ± 4	145 ± 4	133 ± 6	157 ±10	178 ± 12	29 ± 8

administered tyramine in rats. This enhancement of the pressor potency of tyramine by amphetamine is comparable to that reported for monoamine oxidase inhibitors(3). The potentiation was observed following either intravenous or intraduodenal administration of the amphetamine.

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Placental Transport of an Active Enzyme, Invertase.* (31326)

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The majority of studies on placental transport of proteins have involved the transport of antibodies during late stages of fetal development(1). Studies on the uptake of yeast invertase by the liver cell and its concentration in liver lysosomes(2) suggest that invertase might be concentrated in the lysosomes of the rat placenta and from there be transmitted to the fetus. Invertase retains its enzymatic properties for a long period of time within lysosomes of tissues(2) and this activity provides a marker for the presence of the active part of the protein.

Materials and methods. Approximately 2,000 units (μM glucose/min/ml saline) of invertase (Nutritional Biochem.) were injected intravenously in the femoral vein of the pregnant rat. In the first experiment 4 single injections of 1 ml were given at 12-hour intervals, the last injection administered 12 hours prior to autopsy on either day 14 or 15 of pregnancy. Control animals were injected with saline. Homogenates were made

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TABLE I. Invertase Activity in the Fetus, Amniotic Fluid, Yolk Sac, and Placental Labyrinth on Days 14 and 15 of Pregnancy Following 4 Injections of the Enzyme at 12-Hr Intervals with the Last Injection 12 Hr Prior to Autopsy. Results are expressed as units (μM glucose/min) of invertase per unit of tissue (*i.e.*, single fetus, yolk sac, etc.). Mean ± standard error. Numbers in parentheses indicate numbers of rats injected.

Day of pregnancy	Amniotic fluid	Fetus	Yolk sac	Placental labyrinth
14 (5)	.02 ± .00	.05 ± .03	6.45 ± 1.39	4.17 ± 1.24
15 (5)	.07 ± .01	.01 ± .00	16.03 ± 4.30	6.70 ± 1.61
Control (3)	0	0	.01 ± .01	.02 ± .02

TABLE II. Invertase Activity in the Fetus, Amniotic Fluid, Yolk Sac, and Placental Labyrinth on Day 12 of Pregnancy, 2 Hr After Injection of the Enzyme. Results are expressed as units (μM glucose/min) of invertase per tissue unit, *i.e.*, single fetus, etc. Mean ± standard error. Numbers in parentheses indicate numbers of rats injected.

Treatment	Amniotic fluid	Fetus	Yolk sac	Placental labyrinth
Injected (4)	.41 ± .04	.01 ± .01	.29 ± .04	3.50 ± .42
Control (2)	0	0	0	0

of fetuses, yolk sacs, and placental labyrinths in 0.25 M mannitol containing 0.01% triton X-100. Amniotic fluid was collected. Invertase activity was determined by incubation of the samples in 0.25 M sucrose substrate, pH 5.0, for 2 hours. Values obtained by incubation for 10 minutes were subtracted from those for 2-hour incubation and were considered to be "blank" readings. The reactions were stopped by the addition of 0.3 N barium hydroxide and 5% zinc sulfate, and the resultant precipitate was removed by filtering. Glucose in the filtrate was determined by the glucose oxidase reaction.†

The second experiment involved determination of a time curve describing the uptake of invertase in the placental tissues and the transport to the amniotic fluid and fetus. Single injections (1 ml) of invertase were administered on days 13, 14 and 15 of pregnancy. Tissue samples were taken by laparotomy at 2-, 4- and 8-hour intervals and the amount of invertase activity in the various tissue and fluid samples was determined.

An additional experiment consisted of single injections of invertase on day 12 of pregnancy and autopsy 2 hours after injection.

Results and discussion. The data from the multiple injections, Table I, demonstrate that in all animals there was a measurable amount of invertase present in the fetus and amniotic fluid. A large amount of the enzyme was present in the yolk sac and placental labyrinth. No measurable invertase activity was found in control fetuses or amniotic fluid and only a trace in the control yolk sacs and placental labyrinths.

In the 13-day fetus following a single injection, a small but measurable amount of the enzyme was present in every fetal sample and the concentration of enzyme appeared to be stable over the period of time studied (Fig. 1). The amniotic fluid showed a gradual decrease in enzyme activity during the time when the enzyme concentration in the yolk sac was still increasing. The activity in the placenta remained relatively constant over the 8-hour period. Identical time curves were observed on days 14 and 15 of pregnancy.

In 3 of 4 injected animals on day 12 of pregnancy (Table II) invertase was present in small amounts in the fetus 2 hours after injection of the enzyme.

This study has demonstrated that in relatively early stages of pregnancy, an active enzyme can be transported across the placental membranes to the fetus and amniotic fluid. There appears to be a particular concentration of the enzyme in the yolk sac placenta. This agrees with previous indications that the yolk sac may be the means of protein transport across the rodent placenta(3).

Summary. Invertase activity was identified in the rat fetus and amniotic fluid following

† Glucostat special, method 1-A, Worthington Biochemical Corp., Freehold, N. J.

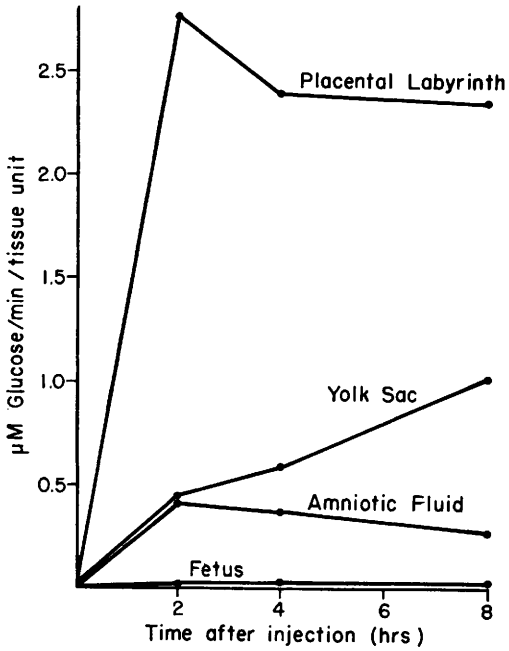

FIG. 1. Invertase activity in fetal and placental tissues at varying time intervals after injection on day 13 of pregnancy. Each point represents a mean value of tissues from 5 injected rats. Tissue unit is a single fetus or part of the placenta. 0 time represents control values.

multiple intravenous injections of the enzyme on days 14 and 15 of pregnancy. Single injections of the enzyme were given on day 13 of pregnancy and samples of conceptuses were taken over an 8-hour time interval. Enzyme analyses showed a low but consistent amount of invertase in the fetus, a gradual decrease in enzyme concentration in the amniotic fluid over the time period studied, a gradual increase in the yolk sac and a relatively stable concentration in the placental labyrinth. Invertase was present in the 12-day fetus 2 hours after injection.

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A Micro Method for Performing Parainfluenza Virus Neutralization Tests.* (31327)

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Tissue culture neutralization tests for the parainfluenza viruses have a variety of applications. These include identification of viral isolates, studies on antigenic relationships between viral strains, determination of the immunogenicity of viral vaccines and serologic diagnosis of human infections (although hemagglutination inhibition (HI) or complement fixation (CF) tests are usually employed for this purpose). Also, neutralization tests are useful in monitoring for antibodies to the parainfluenza viruses in sera of laboratory animals intended for experimental use, or for production of viral immune sera or complement.

Neutralization tests for this group of viruses are generally performed in tube cultures of monkey kidney cells; after inoculation of serum-virus mixtures, the cultures are incubated for an appropriate length of time and the presence of un-neutralized virus determined by testing for hemadsorption of

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