to conclude that the antiviral action of interferon is not through an effect on one of the major synthetic pathways of normal cells. It is also probable that previous results showing effects on normal cells are attributable to impurities in the preparations. While the interferon used in the present experiments is not completely pure, it can be stated that those impurities responsible for effects on normal cells have been eliminated. This explanation is consistent with the results of Baron, Merigan and McKerlie(6) on the effect of crude and purified chick and mouse interferons on growth of cells in tissue culture. These authors clearly distinguish the effects on virus inhibition from the cell growth inhibiting effects of these preparations.

Conclusions. The effect of purified chicken interferon on growth of uninfected chick cells in tissue culture was studied. The criteria for growth used were the increase in amounts of protein, DNA and RNA, and the synthetic rates of protein and RNA. Doses of interferon that were strongly inhibitory to viral growth caused no changes in these parameters.

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Serum Insulin Concentration and Birth Weight in Human Infants.* (30696)

K. SHIMA, S. PRICE AND P. P. FOÀ
Division of Research, Sinai Hospital of Detroit, and Department of Physiology and Pharmacology, Wayne State University School of Medicine, Detroit, Mich.

Transitory hyperglycemia and glycosuria (1), impaired glucose tolerance(2,3) and hyperlipemia(3,4) are signs of the added metabolic burden which pregnancy imposes upon the islands of Langerhans. In predisposed individuals and especially in women who show tendency to bear large infants, this added burden may precipitate the appearance of "metagestational" clinical diabetes(1,3,5). The reasons for the apparent relationship between diabetes and tendency to bear large infants are not clear: a commonly accepted hypothesis states that, in diabetic women, high maternal blood glucose results in fetal hyperglycemia, stimulation of the fetal pancreas, hyperinsulinism and, consequently, increased fetal size. The following observations lend support to this hypothesis: a. the placenta is not an effective barrier against the transfer of glucose and, at least in rabbits, the fetal blood glucose fluctuates with that of the mother, although at slightly lower levels(6); b. the pancreata of large infants and of "normal" infants of diabetic mothers show hypertrophy and hyperplasia of the islets of Langerhans(7-9) and increased insulin content(10); c. diabetic rats tend to produce larger than normal pups with insular hypertrophy and hyperplasia(11-13); d. marked hypoglycemia often occurs in infants of diabetic mothers shortly after birth, when they are separated from the maternal sources of glucose(7); e. increased body fat and carbohydrate content(14,15) and accelerated glucose disposal rate(16) are prevalent in infants of diabetic mothers. Although these observations bear witness to fetal hyperinsulinism, direct measurements of insulin in the blood of the newborn have been few. One group of investigators(17) noted a rise in fetal serum immuno-reactive insulin (IRI) con-

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TABLE I. Effect of 0.01 M EDTA on Recovery of Insulin Added to Human Serum.

	With EDTA			Without EDTA		
Sample	Without added insulin	With added insulin (100 μu)	% recovery	Without added insulin	With added insulin (100 μu)	% recovery
1	67.1	168.0	100.5	140.0	296.5	123.5
2	134.3	265.0	112.5	137.0	339.0	143.0
3	205.0	290.0	95.0	128.5	195.5	85.6
4	231.0	297.0	89.7	135.8	346.5	147.2
5	125.0	245.0	109.0			
6	33.6	160.8	118.0			
7	17.2	128.0	109.0			
8	25.4	148.6	118.5			
9	34.6	140.9	104.7			
Avg ± S.E.			106.3 ± 3.3			124.8 ± 14.1

centration following a rapid intravenous injection of glucose into the mother; other workers(16) found that the rise in blood insulin-like activity (ILA) following administration of glucose, was much larger in the infants of diabetic mothers than in those of non-diabetic mothers, although there were no differences in the fasting state; still other workers found that the ILA(18) and IRI(19) of neonatal blood was significantly higher when the mother was diabetic than when she was normal.

This paper reports data on serum insulin and glucose concentrations in a group of mothers and of newborn infants in relation to the birth weight of the baby.

Materials and methods. The studies were performed on mothers delivered at Sinai Hospital of Detroit and on their infants whose

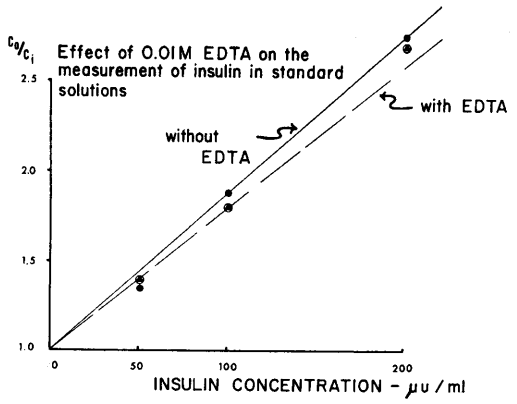

FIG. 1. Insulin standard curve with and without EDTA. Precipitating antibody 1:16; insulin antiserum 1:10,000; ^{131}I-labeled insulin 250 μμg. C_o and C_i are the radioactivities in the insulin-antibody precipitate in absence and presence of unlabeled insulin, respectively(20).

gestational ages ranged from 36 to 41 weeks. All mothers were normal: in particular, there were no cases of diabetes, toxemia or multiple births. Infant blood samples were taken from the umbilical cord vein at time of delivery; maternal blood samples were obtained from the antecubital vein on the morning of the third or fourth day after delivery, before breakfast. The blood was allowed to clot in the refrigerator, centrifuged as soon as possible and the serum was removed and frozen. Immuno-reactive insulin (IRI) was measured in the undiluted serum using the two antibody method of Hales *et al*(20), after inactivation of inhibitors with 0.01 M EDTA(21). This concentration of EDTA does not modify significantly the results obtained with standard insulin solutions (Fig. 1), but improves the recovery of insulin added to human serum (Table I).

Glucagon-free bovine insulin† was used for preparation of the standards and, after mixing with incomplete Freund adjuvant, for preparation of guinea pig anti-insulin serum (22). Since the affinity of human insulin for bovine insulin antibody is not as great as that of bovine insulin itself, the results are expressed as "bovine insulin equivalents" rather than insulin. Blood glucose was measured using the method of Somogyi as modified by Nelson(23).

Results. Fig. 2 shows serum IRI values obtained in 5 groups of infants divided according to birth weight. There were no statistically significant differences between con-

† Gift of Dr. Mary Root, Lilly Research Laboratories.

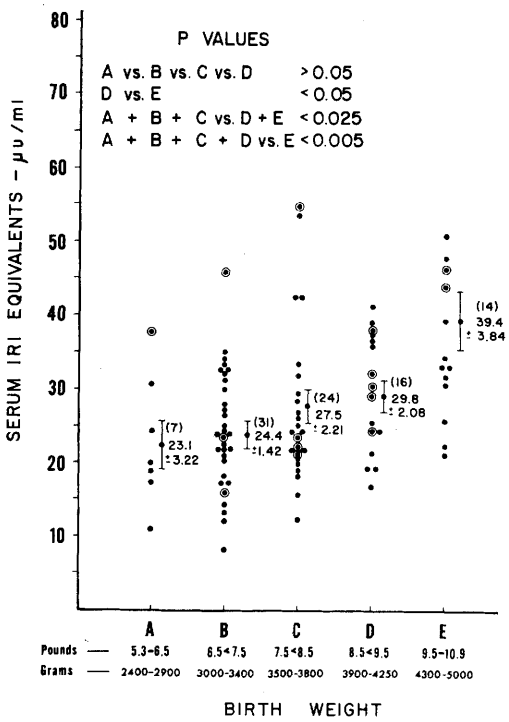

FIG. 2. Serum immuno-reactive insulin in 5 groups of newborn infants divided according to birth weight. Average values ± S.E. indicated. Circles represent cases with family history of diabetes.

secutive groups, except between Groups D and E. When Groups D and E were combined and compared to the combined Groups A, B, and C, the difference and its statistical significance were greater ($P<0.025$; Fig. 2). The greatest difference was obtained when Group E was compared to all other groups as a whole ($P<0.005$; Table II). It will be noted further that all newborn infants weighing more than 10 pounds, except one, had IRI-equivalent values exceeding 30 μu/ml. There were no statistically significant differences between the average blood glucose concentrations of the various groups of infants, regardless of how they were divided (Table II). Similarly, there were no statistically significant differences between the IRI and the blood glucose values of the mothers of large and of normal infants (Table II).

In 47 infants weighing less than 4000 g and in 14 infants weighing 4000 g or more a reliable family history was obtained. Diabetes was present in relatives of 6 (43%) of the 14 "large" babies and in 11 (23.4%) of the 47 "normal" ones. No correlation was found between family history of diabetes and serum glucose and IRI concentrations, either in the mothers or the infants (Table III), although of the 15 infants with family history of diabetes in whom all determinations were made, 10 had either an abnormally high serum IRI or an abnormally high birth weight, or both (Fig. 2).

Discussion. There is no universally accepted definition of "large" infant. Some authors consider abnormally large any newborn baby weighing more than 10 lb or 4.5 kg(24,25); other writers set the normal limit at 9 lb or about 4 kg(2,26). The serum IRI determinations reported here suggest that, although babies weighing 4 kg (9 lb) or more may have an abnormal insular function, the differentiation becomes sharper if one takes a weight of 4.3 kg (9.5 lb) as the dividing line. We were unable to confirm the observation that the plasma insulin concentration of cord blood is lower than that of the maternal venous blood(17), although we used a similar method for determination of insulin and found comparable glucose concentrations in maternal and cord blood. On the contrary, in the case of large infants, we found that IRI concentration was often higher in the serum of the infant than in the serum of the mother, an observation consistent with the idea that the serum insulin of the baby is produced by the baby's own pancreas and that the placenta is an effective barrier to the transfer of insulin(27). No statistically significant correlations were found between infant and maternal serum IRI and their respective blood glucose concentrations. The suggestive correlation between birth weight, serum IRI and family history of diabetes and the statistically significant correlation between birth weight and infant serum IRI, indicate that the excessive size of certain newborn babies may indeed be the result of hyperinsulinism and that, in turn, this hyperinsulinism may be partially the result of genetic factors. This concept may explain why, according to one report (28) prediabetic fathers are also prone to sire overweight infants.

TABLE II. Serum Glucose and Immuno-Reactive Insulin (IRI) in "Normal" and "Large" Newborn Infants and in Their Mothers. The two parts of the table represent the same population using either 4.0 or 4.3 kg as the dividing line.

Group	No.	Serum glucose (mg %, avg ± S.E.)	Serum IRI (μu/ml, avg ± S.E.)	Range
"Normal" (<9 lb or 4 kg)				
Infants	67	78.3 ± 3.17	26.1 ± 1.12*	8.7 — 53.2
Mothers	46	64.8 ± 1.73	27.7 ± 1.15	12.3 — 46.2
"Large" (9 lb or 4 kg or more)				
Infants	25	73.3 ± 4.68	33.9 ± 2.64*	17.3 — 76.8
Mothers	16	69.9 ± 2.71	27.1 ± 2.30	13.1 — 45.1
"Normal" (<9.5 lb or 4. 3 kg)				
Infants	78	76.9 ± 2.75	26.3 ± 1.04†	8.7 — 54.8
Mothers	51	65.5 ± 1.92	28.4 ± 1.14	12.3 — 46.2
"Large" (9.5 lb or 4.3 kg or more)				
Infants	14	72.0 ± 6.64	39.4 ± 3.84†	21.4 — 76.8
Mothers	11	68.6 ± 2.83	25.5 ± 1.48	18.8 — 33.9

* $p < .025$. † $p < .005$.

TABLE III. Relation of Serum Glucose and Immuno-Reactive Insulin (IRI) to Family History of Diabetes.

Group	No.	Serum glucose (mg %, avg ± S.E.)	Serum IRI (μu/ml, avg ± S.E.)	Range
Negative history				
Infant	39	75.6 ± 4.09	27.7 ± 1.63	14.5 — 76.8
Mother	16	68.0 ± 5.48	30.6 ± 2.00	15.9 — 45.3
Positive history				
Infant	15	89.4 ± 5.56	31.7 ± 2.52	16.1 — 54.8
Mother	8	71.0 ± 8.02	31.2 ± 3.41	17.2 — 45.1

The differences between comparable values are not statistically significant.

Summary. Serum immuno-reactive insulin (IRI) and glucose levels were measured in the blood of newborn infants and of their mothers. Infants weighing more than 4 kg at birth were found to have significantly higher serum IRI values than infants of "normal" weight. No statistically significant correlations were found between infant and maternal serum IRI and their respective fasting blood glucose concentrations.

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Stimulation of Detoxification of O-Ethyl O-(4-Nitrophenyl) Phenylphosphonothioate (EPN) by Nikethamide and Phenobarbital.* (30697)

KENNETH P. DUBOIS AND FLORENCE KINOSHITA

Toxicity Laboratory, Department of Pharmacology, University of Chicago, Chicago, Ill.

DiStefano *et al*(1) demonstrated that the acute toxicity of the insecticide, O-ethyl O-(4-nitrophenyl) phenylphosphonothioate (EPN), to rats was reduced to a much greater extent by a combination of nikethamide and atropine than by atropine alone. Rosenberg and Coon(2) found that nikethamide greatly reduces the anticholinesterase action of EPN *in vivo* but the exact mechanism by which this central nervous system stimulant produces its protective effect has remained obscure.

Neal and DuBois(3) recently found that the detoxification of EPN to yield p-nitrophenol involves the action of a microsomal oxidase system in the liver, and developed a quantitative method for measuring the activity of this system. Brazda and Baucum(4) have demonstrated that nikethamide induces synthesis of a microsomal enzyme that catalyzes the oxidation of pentobarbital. It thus seemed possible that the protective action of nikethamide against EPN poisoning might involve induced synthesis of the detoxification system for EPN. The results of the present study demonstrated that treatment of rats with nikethamide increases the ability of their livers to detoxify EPN *in vitro*. Phenobarbital, a known stimulant of microsomal enzyme induction(5), also increased activity of the detoxification system and reduced the acute toxicity of EPN to rats.

Materials and methods. Adult, male and female Sprague-Dawley rats were used. EPN was kindly supplied by the E. I. duPont Co., Wilmington, Del. For administration to animals, EPN and nikethamide were dissolved in 20% ethanol and 80% propylene glycol. Sodium phenobarbital was dissolved in water. All drugs were given by the intraperitoneal route and the concentrations of solutions were adjusted so that the animals received amounts equivalent to 0.1% of the body weight. The doses of nikethamide and phenobarbital used for enzyme induction were 100 mg/kg/day and 50 mg/kg/day respectively. The rate of degradation of EPN to p-nitrophenol was measured by the method of Neal and DuBois (3) using whole liver homogenates. LD_{50} values were calculated from the 14-day mor-

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