Insulin and Free Fatty Acid Levels During Oral Glucose Tolerance Tests and Their Relation to Age in 70 Healthy Children

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SUMMARY

In 70 healthy children aged from three months to 15 years, blood glucose, immunoreactive insulin, and free fatty acids were measured during a three-hour glucose tolerance test. The results are presented for the whole group as well as for three age groups: three months-five years, six-10 years, and 11-15 years.

It is demonstrated that (1) glucose levels are significantly lower in young children (younger than five years); (2) there are no significant age-related changes in free fatty acid concentrations; (3) insulin levels are increasing constantly and significantly with age, the most strikingly at the age of onset of puberty; the absence of notable changes in glucose tolerance results in a rise of the I/G ratio as well.

The causes for the increase of insulin secretion with age, whether of peripheral or pancreatic order, are still speculative. DIABETES *25*:505-08, June, 1976.

Plasma insulin and free fatty acid responses to glucose may be difficult to interpret in children, one difficulty being the poor availability of reference values. Indeed, there are few investigations carried out in a sufficient number of subjects to allow statistical analysis,[1-3] and the values of several other studies are limited by the small number of children.[4-7]

The purpose of the present study was to contribute to a better assessment of normal insulin and free fatty acid responses to glucose in children of different ages, particularly as marked differences in glucose tolerance at different stages of childhood have already been reported.[8]

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Accepted for publication February 26, 1976.

SUBJECTS AND METHODS

In 70 healthy, normal-weight children without family history of diabetes who were hospitalized for and recovering from nonmetabolic diseases, a standard oral glucose tolerance test was carried out following the recommendations of the World Health Organization* with 30 gm. of glucose per m^2 of body surface and starting at 9:00 a.m. after an overnight fast. They were 41 boys and 29 girls, and ages ranged from three months to 15 years (mean age 8.8 years). For analysis of age changes, the children were divided into three arbitrarily chosen age groups of five years each, i.e. group I: three months to five years (16 subjects), group II: six to 10 years (21 subjects), and group III: 11 to 15 years (33 subjects).

Blood glucose, immunoreactive insulin (I.R.I.), and free fatty acids (F.F.A.) in venous plasma were measured during fasting and three hours following glucose ingestion. Blood glucose was determined by the method of Nelson-Somogyi,[9] I.R.I. by the radioimmunoassay of Yalow and Berson,[10] and F.F.A. by the technic of Duncombe.[11]

RESULTS

The mean values for blood glucose, I.R.I., and F.F.A. are presented in table 1 for the total number of subjects and each age group.

Mean blood glucose values were significantly higher in groups II and III than in group I at 0, 30, and 60 minutes.

No changes were noticed in the F.F.A. levels of the different age groups. Though the levels were slightly

*W.H.O. Series of technical reports no. 310. Le diabète sucré, rapport d'un comité d'experts, Genève, 1965.

TABLE 1

Oral glucose tolerance test in 70 normal children
Means ± S.E.M.

	Time (minutes)	0	30	60	120	180
	Blood glucose	81	117	105	86	73
	mg./100 ml.	±4.0	±5.1	±5.0	±4.7	±4.3
Total	I.R.I.	8.4	50.4	43.5	22.5	10.4
n=70	μU./ml.	±3.3	±6.4	±5.9	±4.8	±3.6
	F.F.A.	427	249	134	95	265
	μEq./L.	±14.6	±11.0	±7.7	±7.1	±13.1
	Blood glucose	71*‡	100*‡	88*‡	82	69
	mg./100 ml.	±3.7	±4.3	±3.8	±4.3	±4.2
Group I	I.R.I.	5.8	27.4*‡	24.8‡	16.1‡	8.2
n=16	μU./ml.	±2.3	±4.0	±4.1	±3.8	±2.9
	F.F.A.	478	233	167	95	371
	μEq./L.	±14.3	±9.9	±8.1	±7.5	±11.5
	Blood glucose	83*	122*	105*	83	74
	mg./100 ml.	±3.9	±4.7	±4.9	±4.5	±4.2
Group II	I.R.I.	6.0	46.5*	36.1†	16.9†	5.4†
n=21	μU./ml.	±2.4	±5.4	±4.5	±4.5	±3.0
	F.F.A.	423	260	134	107	330
	μEq./L.	±14.4	±11.2	±8.8	±8.0	±14.4
	Blood glucose	85‡	123‡	114‡	89	73
	mg./100 ml.	±4.1	±5.3	±5.4	±5.2	±4.8
Group III	I.R.I.	11.3	64.0‡	57.3†‡	29.0†‡	14.4†
n=33	μU./ml.	±3.7	±6.9	±6.2	±4.6	±4.2
	F.F.A.	419	246	128	89	217
	μEq./L.	±18.3	±11.6	±7.3	±6.4	±13.1

Significant differences ($p<0.01$)
Group I versus II (*)
Group II versus III (†)
Group I versus III (‡)

		Insulinogenic index				
Total		0.10	0.42	0.42	0.27	0.14
Group I	I/G	0.08	0.26	0.28	0.48	0.09
Group II		0.08	0.39	0.36	0.23	0.08
Group III		0.13	0.51	0.52	0.34	0.20

higher in the younger children, the differences were not significant.

Insulin secretion, particularly poststimulative, was found to increase progressively with age, the most important rise being noted between groups II and III. The differences are significant between groups I and II at 30 minutes, between groups II and III at 60, 120, and 180 minutes, and between groups I and III at all times except 0. This elevation is not attributable to the moderate rise of blood glucose, for the insulinogenic index (I/G ratio) as well increases constantly with age (table 1).

Glucose values and insulin responses were weakly but significantly related, but only at 30 minutes. Figure 1 shows the distribution of individual insulin responses as a function of blood glucose levels at 30 minutes in all age groups. A significant correlation is observed in groups I and III; however, the insulin responses to corresponding glucose values were higher in group III than in group I.

The progressive elevation with age of insulin responses in relation to glucose stimulus is shown again in figure 2, where the difference between fasting and 30-minute insulin values (Δ insulin) are plotted against the Δ of glucose. There is a significant correlation in all three age groups, and the capacity of blood glucose to stimulate insulin secretion rises from one age group to the other.

DISCUSSION

In a previous study on the glucose tolerance of 300 healthy children[8] we reported a significant difference between the glucose levels of children under and above three years of age, observed by Grant[5] as well. Our present data are quite similar, though the age groups chosen in this series are slightly different. Actually, because of the limited availability of very young control subjects, only seven children were less than three years old. In order to obtain age groups approximately balanced with respect to the number of subjects and

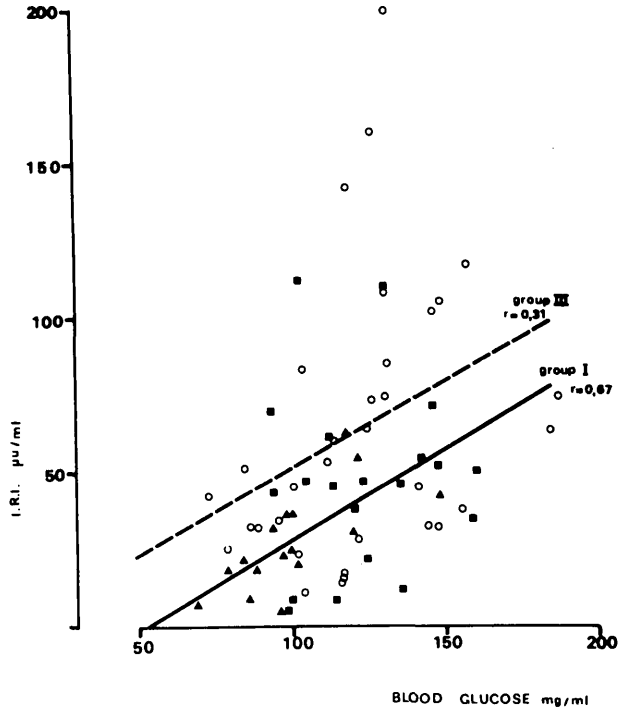

FIG. 1. Distribution of individual insulin values as a function of blood glucose values 30 minutes postabsorptive, and regression lines of groups I and III (no correlation in group II). ▲ group I, ■ group II, O group III

the age span, we enlarged group I to include children up to five years old.

Cole and Epel[1] found lower blood glucose values only in infants of younger than 18 months. Though we believe such a difference very likely, we were unable to support it by our present study. Only three of our children were that young (they were aged three months, seven months, and 18 months), and their data did not differ from those of the elder children of group I.

Rosenbloom et al.[3] did not find significant age differences in blood glucose in a large series of normal children.

We observed an average fasting F.F.A. level a little lower than that described by other authors,[6,7,12] but the relative changes following glucose absorption are similar.

The analysis of insulin levels shows a marked increase of insulin secretion with age, already reported by other authors. Table 2 presents a comparison of the results of several investigators who measured insulin levels during glucose tolerance tests in children. The results of Grant[5] are comparable to ours. Rosenbloom et al.[3] found highly significant age changes in all three groups of children corresponding to our age groups and at all times, but his values are almost twice those obtained by our study. The higher glucose load administered in this study (1.75 gm./kg. body weight, which is about twice the amount based on body surface given in our laboratory) might partly explain this difference. Varying glucose loads have been shown to affect insulin responses more significantly than glucose levels by Castro et al.[13] But the fasting insulin levels they found were higher than ours as well. Moreover, all the other authors quoted, except Grant,[5] used the same dosage of glucose, i.e. 1.75 gm./kg., yet their results are comparable to ours. Parker et al.[6] and Cole and Epel,[1] on the other hand, obtained lower peak values at 30 minutes, and the latter authors, when studying plasma insulin in different age groups, did not find any age-related difference.

The age-related increase of absolute insulin secretion is paralleled by a rise of the I/G ratio, as Rosenbloom et al.[3] also found. As demonstrated in figures 1 and 2, poststimulative blood-glucose levels and insulin secretion are related, and this relation is influenced by age. At present, one might only speculate about the reasons why insulin secretion increases with age. Is it a sign of insulin resistance due to diminished insulin binding to membranous receptors with age? This phenomenon in adult chemical diabetes and obesity[14,15] might be a symptom of aging as well, but at present there is no evidence for this in the child. Another hypothesis would be a growing insulin/glucagon ratio as a consequence of maturation. Recent studies have shown that a close interrelationship between alpha- and beta-cell function and a constantly changing insulin-glucagon balance play an important role in regulating energy metabolism.[16]

The demonstration in figure 2 of a direct modification, by age, of the insulin response induced by a

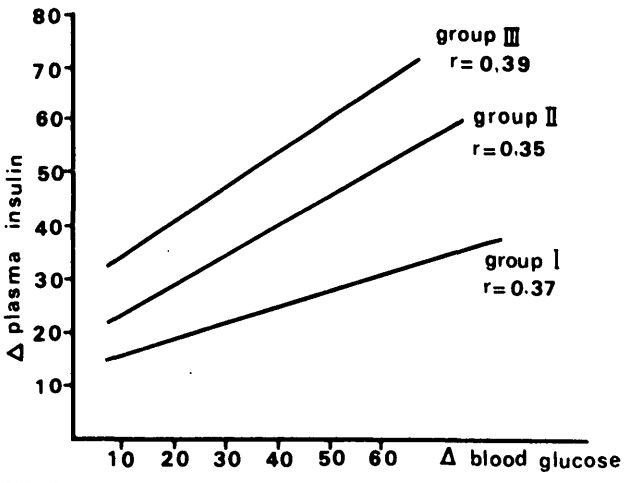

FIG. 2. Correlation between Δ values of glucose and insulin, Δ being the difference between 30-minute and fasting values. Regression lines of groups I, II, and III.

TABLE 2

Published mean plasma insulin values (μU./ml.) during O.G.T.T. in normal children

Author	No. of subjects	Age (years)	0	30	60	120	180 minutes
Cole and Epel	112	1½-12	9.6	29.3	32.5	21.0	17.0
Martin and Martin	30	2-16	6	50	43	37	16
Parker et al.	10	1-13	10	30	47	25	14
Grant	12	<3	4	25	20	15	
	6	>3	8	62	65	12	
Rosenbloom	33	1½-5	10.6	47.6	40.7	30.5	14.0
	37	6-10	14.9	80.0	57.9	49.8	13.5
	31	>11	20.1	115.2	97.2	81.1	43.5
Authors	70	0-15	8.4	50.4	43.5	22.5	10.4
	16	0-5	5.8	27.4	24.8	16.1	8.2
	21	6-10	6.0	46.5	36.1	16.9	5.4
	33	11-15	11.3	64.0	57.3	29.0	14.4

given rise of the blood glucose level favors a growing beta-cell sensitivity to glucose stimulus with age.

The striking insulin changes between groups II and III suggest that the influence of the over-all hormonal changes occurs with puberty. However, as in this study we had not divided the children according to sexual maturity, we were not able to establish any correlation between puberty and insulin levels. This question will have to be precised subsequently.

Because of a usually early puberty in girls and of their different body composition (particularly thicker subcutaneous adipose tissue) one would expect higher insulin levels in girls than in boys of this age group. We actually found mean insulin values at 30 and 60 minutes about 25 per cent lower in the boys of group III than in the girls, but this difference was not significant. On the other hand, the same difference in insulin responses between boys and girls was observed in group II, aged six to 10 years; only the boys and girls of group I had identical insulin levels. Analysis of a larger series of adolescent and preadolescent children will have to be made to confirm the existence of sex-related insulin secretion in childhood.

Our results confirm the well-known wide range of individual insulin responses, which sometimes makes the interpretation of borderline values difficult. Indeed, the theoretical limits of normal insulin secretion, especially the lower limit, as defined by two standard deviations, are very wide and not always of practical use. For diagnosis, the consideration of the insulin response in relation to the glucose tolerance curve will be decisive in most cases.

Standard values for insulin secretion in children are by themselves of great theoretical value, however, as they form a base of reference necessary for further investigations.

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