Metabolism of Oral Glucose in Children Born Small for Gestational Age: Evidence for an Impaired Whole Body Glucose Oxidation

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Epidemiological studies indicate that intrauterine growth restriction confers an increased risk of developing type 2 diabetes mellitus in subsequent life. Several studies have further documented the presence of insulin resistance in young adults or adolescent children born small for gestational age. Since most studies addressed postpubertal individuals, and since puberty markedly affects energy metabolism, we evaluated the disposal of oral glucose in a group including mainly prepubertal and early pubertal children with intrauterine growth restriction and in healthy age- and weight-matched control children. All children had an evaluation of their body composition by skinfold thickness measurements. They were then studied in standardized conditions and received 4 consecutive hourly loads of 180 mg glucose/kg body weight to reach a near steady state. Energy expenditure and substrate oxidation were evaluated during the fourth hour by indirect calorimetry. Compared to both age- and weight-matched children, children born small for gestational age had lower stature. Their energy expenditure was not significantly decreased, but they had lower glucose oxidation rates. These results indicate that metabolic alterations are present early in children born small for gestational age, and are possibly related to alterations of body composition.

LARGE EPIDEMIOLOGICAL studies have shown that type 2 diabetes, high blood pressure, and atherosclerotic disease are more prevalent in adults who were born small for their gestational age.[1-3] This led to the hypothesis that a shortage in nutrients during the gestational period triggers the development of a thrifty phenotype.[4,5] According to this hypothesis, children and adults with such a thrifty phenotype develop metabolic diseases when subsequently exposed to nutrient overload in postnatal life. In support of this hypothesis, several animal studies have documented that energy or protein restriction during fetal development leads to a decreased β-cell mass and insulin secretory capacity,[6] muscle and adipose tissue insulin resistance,[7,8] and increased hepatic glucose production.[5,9,10] These observations are made several weeks postnatally, suggesting that additional factors play a role in the alterations induced during fetal development. In humans, the hormonal changes associated with puberty are known to alter significantly insulin actions and glucose homeostasis[11] and hence may enhance the alterations of glucose homeostasis induced by fetal undernutrition.

In young adults or adolescent humans born small for gestational age, it was demonstrated that insulin-mediated glucose uptake was on average lower than in individuals with normal birthweight.[12,13] However, there is little information regarding the metabolic alterations associated with intrauterine growth restriction in prepubertal and early pubertal children. We therefore assessed, in a group of children aged 8 to 14 years, including mainly prepubertal children and children in early pubertal stages, the metabolic fate of ingested glucose and compared the results in 2 groups of children without intrauterine growth restriction. One group consisted of children matched for their age; the other group consisted of children matched for their current weight. The results obtained indicate that intrauterine growth restriction is associated with higher adiposity and impaired glucose oxidation after glucose ingestion already before puberty.

MATERIALS AND METHODS

Selection and Inclusion of Subjects

Fifteen children and adolescents aged 12.1 ± 2.0 years (range, 8 to 14 years) who were born small for gestational age participated in this study. The criteria for inclusion were a birthweight and height for gestational age less than percentile 10. Children and adolescents were among patients referred to the Endocrinology and Diabetology Unit of the Children's Hospital, Lausanne, Switzerland for growth restriction. Twenty-four healthy children (11.6 ± 2.1 years ; range, 8 to 14 years) born with a normal weight were recruited in the schools of the city of Lausanne. Recruitment was done by sending a circular letter to parents, inviting them to an information session. Fifteen healthy subjects were selected to match the ages and 15 other subjects were selected to match the weights of the children born small for gestational age. These healthy children had previously been included in a study addressing the effect of age on whole body glycogen turnover.[14] Written consent was obtained from the parents and oral consent from all children. Exclusion criteria included obesity (defined as weight for height > 97th centile), any known disease, treatment with growth hormone (past or present), gestational diabetes in the mother, and type 1 diabetes in first-degree relatives. The pubertal stage (Tanner stage) was assessed based on breast and pubic hair development in girls and on external genitalia development in boys.[15,16] The experimental protocol was approved by the Ethical Committee of Lausanne University, School of Medicine.

Experimental Protocol

All experiments were performed between 8 AM and noon. Subjects had consumed a light breakfast consisting in a cup of tea with skimmed milk and 10 g sugar and 2 crackers at 6:30 AM. After anthropometric measurements were obtained, children were maintained in a bed in a semirecumbent position and remained quiet while watching movies on a video until the end of the experiment. At 8 AM, 9 AM, 10 AM, and 11 AM, they drank a solution of 20% dextrose, to provide each hour 180 mg glucose/kg body weight. We have recently documented, in healthy

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Table 1. Anthropometric Characteristics

	n	Age (yr)	Gender	Weight (kg)	Height (cm)	Fat-Free Mass (kg)	Fat Mass (kg)	Fat Mass (% total body weight)
Children born small for gestational age	15	12.1 ± 2.0	7 F/8 M	35.1 ± 11.0	136.8 ± 10.3	27.7 ± 7.1	7.5 ± 4.3	19.8 ± 5.8
Age-matched children with normal birthweight	15	12.0 ± 2.1	6 F/9 M	41.8 ± 12.9g*	150.8 ± 16.0*	35.0 ± 10.6*	6.8 ± 3.2	16.0 ± 4.5
Weight-matched children with normal birthweight	15	10.9 ± 2.0*	6 F/9 M	35.6 ± 10.2	143.2 ± 12.9*	29.9 ± 8.0*	5.6 ± 2.5*	15.2 ± 3.8*

NOTE. Values are mean ± 1 SD.

*$P < .05$ or less *v* children born small for gestational age.

adults, that such repeated administration of small glucose loads allows to reach a near steady state after 3 to 4 hours, corresponding to the period when the measurements were performed.[17]

Between 10 AM and noon, respiratory gas exchanges were continuously monitored with a deltatrac II (Datex Instruments, Helsinki, Finland), using a ventilated hood. The measurements were briefly interrupted at 11 AM for ingestion of the fourth glucose load. The measurements obtained between 10 AM and 11 AM were discarded to allow the children to get accustomed to the monitoring procedure. Respiratory exchanges measured between 11 AM and noon were used for calculation of energy expenditure and net substrate oxidation rates[18]: net glucose oxidation (mg/min) = $4.53 \cdot V_{CO_2} - 3.18 \cdot V_{O_2} - 3.6 \cdot N$; net lipid oxidation (mg/min) = $1.61 \cdot V_{O_2} - 1.61 \cdot V_{CO_2} - 1.8 \cdot N$; and net protein oxidation (mg/min) = $6.25 \cdot N$, where V_{O_2} and V_{CO_2} are in milliliters per minute and N is urinary nitrogen excretion in milligrams per minute.

At 11:30 AM, ie, 30 minutes after administration of the last glucose load, a capillary blood sample was obtained at the fingertip and blood glucose concentration was measured with a Precision Q1D glucometer (Medisense; Abbott AG, Baar, Switzerland).

A timed urine collection was obtained between 8 AM and noon to calculate net protein oxidation from urinary urea excretion.

Anthropometric Measurements

On the day of the study, the following anthropometric parameters were obtained: height, body weight, and skinfold thickness measured using a Holtain skinfold caliper at the biceps, triceps, subscapular, and suprailiac sites. All skinfold thickness measurements were done in triplicate to the nearest millimeter by the same investigators (F.J. and R.S.). Fat mass and fat-free mass were calculated from skinfold thickness values using the Deurenberg formulas.[19] These data are presented in Table 1.

Statistical Analysis

All results in the text and tables are expressed as mean ± 1 SD. Comparison of means between the children born with intrauterine growth restriction and the 2 groups of age- or weight-matched children born with normal weight was done with analysis of variance (ANOVA) and post hoc tests.

RESULTS

Anthropometric Parameters

At birth, both weight and height of children born small for gestational age and control subjects were significantly different (weight: 2.4 ± 0.4 *v* 3.1 ± 0.1 kg ; height: 43.7 ± 0.8 *v* 49.0 ± 0.4 cm; $P < .001$ in both cases), indicating symmetrical intrauterine growth restriction in children born small for gestational age. Compared to age-matched children, children born small for their gestational age had a smaller stature and weighed 6.1 kg less on average. Their percent body fat was 10% higher ($P = .08$), whereas their fat-free mass was 21% lower ($P < .002$). Children born small for gestational age were also compared to a group of weight-matched children. This group of children was on average 1.2 years younger. However, children born small for gestational age remained significantly shorter, and had a higher percent body fat ($P < .01$) and a higher body fat mass ($P < .05$) compared to this latter group of younger, weight-matched children (Table 1).

The children included in each group were aged 8 to 14 years, and hence encompassed several stages of pubertal development. The distribution of pubertal stages was identical in the group of children born small for gestational age (7 P1, 4 P2-P3, 4 P4-P5) and in the age-matched control group (6 P1, 4 P2-P3, 5 P4-P5). Younger children included in the weight-matched control group had on average lees advanced pubertal developments (8 P1, 5 P2-P3, 2 P4-P5).

Energy Metabolism Glucose Oxidation and Glycemia

Table 2 shows the energy expenditure and net substrate oxidation rates measured during the fourth hour of hourly oral glucose ingestion. Total energy expenditure was not significantly different between children born small for gestational age and age- or weight-matched control groups. When expressed relative to body weight, children born small for gestational age tended to have a 8% lower energy expenditure compared to weight-matched children with normal birthweight, but the difference did not reach significance ($P = .09$). Net glucose oxidation was significantly lower and net lipid oxidation was higher in children born small for gestational age when compared to both age- and weight-matched control groups. Net carbohydrate oxidation represented 61.1% of total energy expenditure in children born small for gestational age verus 77.1% in age-matched ($P < .002$) and 75% in weight-matched controls ($P < .05$). This difference in glucose oxidation remained when data were normalized for fat-free mass to account for differences in body composition (children born small for gestational age: 4.4 ± 0.3 mg/kg fat-free mass versus 5.4 ± 0.3 in weight-matched and 5.0 ± 0.2 in age-matched children ($P < .05$ in both cases). Conversely, net lipid oxidation was higher in children born small for gestational age than in age- and weight-matched children with normal birthweight. The groups of age-

Table 2. Energy Metabolism

	Energy Expenditure (kcal/kg/min)	Net Glucose Oxidation (mg/kg/min)	Net Glucose Oxidation (% energy expenditure)	Net Lipid Oxidation (mg/kg/min)	Net Lipid Oxidation (% total energy expenditure)
Children born small for gestational age	0.022 ± 0.05	3.55 ± 0.94	61.1 ± 15.6	0.55 ± 0.38	21.0 ± 14.8
Age-matched children with normal birthweight	0.021 ± 0.05	4.21 ± 0.70*	77.1 ± 12.7*	0.16 ± 0.30*	6.0 ± 12.4*
Weight-matched children with normal birthweight	0.024 ± 0.005	4.60 ± 0.89*	75.0 ± 18.0*	0.26 ± 0.48	8.2 ± 17.9*

NOTE. Values are mean ± 1 SD.

*$P < .05$ or less *v* children born small for gestational age.

matched and weight-matched children did not differ significantly from each other regarding all metabolic parameters collected. Capillary glucose concentrations measured 30 minutes after ingestion of the last glucose load were 6.6 ± 1.5 mmol/L in children born small for gestational age versus 6.2 ± 0.6 and 6.3 ± 0.80 mmol/L in age- and weight-matched children, respectively (difference not significant).

DISCUSSION

Epidemiological studies have clearly documented that children born small for gestational age are at increased risk of developing type 2 diabetes as adults.[1-3] The individuals included in these studies were 40 years of age or older, suggesting that the occurrence of diabetes is a late event. Other studies, performed in small groups of young adults or adolescents have documented that individuals born small for gestational age have on average a lower insulin-mediated glucose disposal, suggesting a lesser degree of insulin sensitivity in this subset of patients.[12,13] The vast majority of studies published reported on postpubertal children. However, the hormonal changes that occur during puberty markedly affect energy and glucose metabolism[11] and may have amplified the alterations present in these children.

In the present study, we have documented the metabolic fate of exogenous, orally administered glucose in a group of children born small for gestational age during prepubertal and early pubertal stages of development. Weight and height were symmetrically reduced in this group of children. Furthermore, there was no indication in the medical record and family history of these children for other causes of low birthweight. We therefore assumed that it was due to intrauterine growth restriction. However, since the criteria used to detect children small for gestational age is a birthweight below the 10th percentile, it is statistically possible that some children without intrauterine growth restriction were included as well.[20]

Children born small for gestational age had a lower glucose oxidation rate than children of the same age with normal birthweight. Since children born small for gestational age were significantly lighter than normal children of the same age, we also compared their results with those obtained in a group of younger children matched for weight. Children born small for gestational age had lower glucose oxidation when compared to this latter, weight-matched control group too, even though lean body mass did not differ. This clearly indicates that stimulation of glucose utilization in lean tissues was impaired in children born small for gestational age.

The data obtained in this study do not allow to determine which tissues or organs oxidized less than normal glucose in children born small for gestational age. It has been demonstrated, in adults, that skeletal muscle was responsible for the major portion of insulin-mediated glucose metabolism[21,22] and hence appears as a prime candidate for the alterations of glucose metabolism that occur in individuals having suffered intrauterine growth restriction. Jaquet et al[12] have indeed documented that insulin regulation of gene expression was altered in young adults born small for gestational age. Although this does not provide an explanation for the mechanism of insulin resistance, it nonetheless documents that muscle insulin responsiveness is likely to be permanently or at least durable affected by fetal undernutrition.

Children born small for gestational age had a significant restriction of statural growth. They also had less lean body mass, and fat represented a higher percentage of their total weight than normal children. Similar observations have been reported by other investigators.[23] We therefore considered the possibility that their lower glucose oxidation was merely secondary to a reduction in lean body mass. This explanation appears unlikely since the differences remained significant when data were re-expressed normalized for fat-free mass. No blood samples were obtained in these groups of young children and we have therefore no direct information on plasma glucose, insulin, and fatty acid levels during the experimental procedure. It is tempting, however, to speculate that the increased fat mass of children born small for gestational age was associated with a lesser suppression of plasma free fatty acid concentrations after oral glucose. The high fatty acid levels would in turn be responsible for insulin resistance and a low glucose oxidation.[24,25] Features consistent with this hypothesis have indeed been reported by Jaquet et al[12]; these investigators performed clamp studies and observed an impaired suppression of plasma free fatty acids by hyperinsulinemia in young adults born small for gestational age. Other investigators, however, failed to observe a direct relationship between blood lipids and insulin resistance in adults having suffered fetal undernutrition.[26]

It is not possible, based on the present data, to pinpoint the mechanism responsible for the alteration of postprandial glucose metabolism in children born small for gestational age.

Based on reports from the literature, it is, however, possible to propose that hormonal dysregulation may be responsible for both changes in body composition and insulin resistance. The general picture presented by these children, ie, small stature, increased fat mass, and features consistent with insulin resistance are highly reminiscent of a syndrome of growth hormone deficiency. In both children and adults, growth hormone deficiency is associated with a gain in fat mass.[27] Furthermore, substitutive treatment of growth hormone–deficient adults leads to a reduction in body fat together with an improvement of insulin sensitivity.[28,29] Due to their small stature, all of our children born small for gestational age had an endocrine evaluation. None was found to be growth hormone–deficient. Several investigators proposed that fetal undernutrition may lead to a state of growth hormone resistance in postnatal life.[30-32] This hypothesis was supported by the clinical observation that children born small for gestational age required higher doses of growth hormone to stimulate growth than children with small stature not associated with intrauterine growth restriction.[33,34] Alternatively, it has been observed, in animal models, that undernutrition during pregnancy produces a decreased expression and activity of the enzyme 11β-hydroxysteroid dehydrogenase type 2 in placenta and fetal tissues.[35,36] This enzyme is responsible for the local conversion of cortisol into inactive cortisone. Furthermore, it has been recently observed in mice that overexpression in adipocytes of 11β-hydroxysteroid dehydrogenase type 1, which catalyzes the reverse reaction, ie, the conversion of cortisone into cortisol leads to the development of obesity and a syndrome of insulin resistance presumably through enhanced cortisol concentrations.[37,38] It is therefore tempting to speculate that persistant alterations of these enzymes in children born small for gestational age may contribute to changes in both body composition and insulin sensitivity. At this stage, the hypothesis that dysregulation of the somatotrophic and/or pituitary adrenal axis might be responsible for both a small stature and the development of insulin resistance secondary to changes in body fat content appear certainly attractive, but remains to be further evaluated.

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