Prothrombotic State, Cardiovascular, and Metabolic Syndrome Risk Factors in Prepubertal Children Born Large for Gestational Age

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OBJECTIVE — To evaluate metabolic syndrome and cardiovascular disease risk factors in prepubertal children born large for gestational age (LGA) to nondiabetic, nonobese mothers.

RESEARCH DESIGN AND METHODS — At 6–7 years of age, the comparison of various factors was made between 31 LGA and 34 appropriate-for-gestational-age (AGA) children: fibrinogen, antithrombin III, protein C and S, fasting insulin, glucose, homeostasis assessment model of insulin resistance (HOMA-IR) index, adiponectin, leptin, visfatin, IGF-1, IGF-binding protein (IGFBP)-1, IGFBP-3, lipids, and the genetic factors V Leiden G1691A mutation, prothrombin 20210A/G polymorphism, and mutation in the enzyme 5,10-methylenetetrahydrofolate-reductase gene (MTHFR-C677T).

RESULTS — LGA children had higher levels of leptin ($P < 0.01$), fasting insulin ($P < 0.01$), and HOMA-IR ($P < 0.01$), but lower IGFBP-3 ($P = 0.0001$), fibrinogen ($P = 0.0001$), and lipoprotein(a) ($P < 0.001$) than AGA children. Significantly more LGA children were homozygous for the MTHFR-C677T mutation ($P = 0.0016$).

CONCLUSIONS — Being born LGA to nondiabetic, nonobese mothers is associated with diverse effects on cardiometabolic risk factors at prepuberty.

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Large-for-gestational-age (LGA) infants may be at risk for the development of obesity and insulin resistance (1–4). A relationship between excess birth weight and metabolic syndrome (MetS) and cardiovascular disease (CVD) risk factors (1–4) has not yet been clearly demonstrated.

The aim of this study was to evaluate markers of the prothrombotic state and other MetS and CVD risk factors in prepubertal children born LGA to nondiabetic, nonobese mothers.

RESEARCH DESIGN AND METHODS

The study group consisted of 64 singleton Caucasian children, born at term: 31 (10 female, 21 male) were LGA (birth weight ≥95th percentile for gestational age), and 33 (12 female, 21 male) appropriate for gestational age (AGA) (birth weight 10th–90th percentile). No mother was obese (pregestational BMI ≥30 kg/m^2), and all had a normal glucose challenge test during pregnancy.

The children were examined at 6–7 years of age, at which time all were prepubertal. The data compared were: obesity indexes [waist circumference, body weight, and BMI]; arterial blood pressure (BP) expressed as *z* scores; blood levels of fibrinogen, antithrombin III, protein C and S, lipoprotein(a) [Lp(a)], adiponectin, leptin, visfatin, IGF-1, IGF-binding protein (IGFBP)-1, IGFBP-3, fasting insulin (I_F) and glucose (G_F) levels, the homeostasis assessment model of insulin resistance (HOMA-IR) index, the lipid profile, and the genetic factors V Leiden G1691A mutation, prothrombin 20210A/G polymorphism, and mutation in the enzyme 5,10-methylenetetrahydrofolate-reductase gene (MTHFR-C677T). Venous blood samples were drawn from the children after a 12-h overnight fast. Written informed parental consent was obtained for the participation of each child, and the study was approved by the local research ethics committee.

Components of the IGFs-axis, leptin, and adiponectin levels were measured by ELISA. Serum visfatin COOH-terminal levels were determined by a competitive enzyme immunoassay (Phoenix Pharmaceuticals). Coagulation assessment was determined using functional methods and chromatometric assays. Genomic DNA was isolated from the leukocytes of peripheral whole-blood samples collected in EDTA-anticoagulant according to standard methods (5). Lipids and I_F and G_F levels were determined with techniques previously described (1).

Data were analyzed by ANOVA and multiple regression analysis using the StatView software package of SAS Institute (Cary, NC).

RESULTS

The anthropometric and laboratory findings are depicted in Table 1. The significant differences in leptin, I_F, and HOMA-IR between LGA and AGA children persisted after controlling for age, sex, and BMI.

Homozygocity for the MTHFR-C677T mutation was detected in 12 LGA and 2 AGA children ($P = 0.002$) and heterozygocity in 19 LGA and 8 AGA children ($P = 0.003$). Three LGA and none of the AGA children were het-

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Table 1—*Characteristics, anthropometric indices, and indices of the prothrombotic state and insulin resistance, components of the IGFs-axis, lipid profile, and adipocytokines (means ± SD) at prepuberty of children born LGA (birth weight ≥95th percentile) or AGA (birth weight 10th–90th percentile)*

Characteristics and parameters	LGA group	AGA group	*P* value
n	31	33	—
Age (years)	6.5 ± 0.5	6.4 ± 0.6	ns
Body weight (kg)	32 ± 8	24 ± 6	<0.01
Body height (cm)	126 ± 8	119 ± 9	0.08
Waist circumference *z* score	0.80 ± 0.98	0.06 ± 1.3	0.05
BMI *z* score	0.80 ± 0.80	−0.20 ± 0.8	<0.001
Systolic BP *z* score	0.49 ± 0.41	0.41 ± 0.43	ns
Diastolic BP *z* score	0.71 ± 0.35	0.51 ± 0.5	ns
Prothrombin time (s)	13.29 ± 0.53	13.2 ± 0.52	ns
APTT (s)	37.04 ± 22	38.64 ± 24	ns
Fibrinogen (μmol/l)	7.67 ± 0.9	10.05 ± 2.3	≤0.0001
Antithrombin III (%)	104 ± 50	107 ± 69	ns
Protein C (%)	99 ± 11	97 ± 18	ns
Protein S (%)	69 ± 24	70 ± 19	ns
Fasting glucose (mmol/l)	5.1 ± 0.5	4.9 ± 0.6	ns
Fasting insulin (pmol/l)	48.6 ± 20.1	27 ± 24.3	<0.01
FGIR	0.11 ± 0.05	0.25 ± 0.11	≤0.0001
HOMA-IR	1.5 ± 0.6	0.8 ± 0.7	<0.01
IGF-1 (μg/l)	189 ± 115	140 ± 84	0.06
IGFBP-1 (μg/l)	84 ± 33	88 ± 31	ns
IGFBP-3 (mg/l)	2.6 ± 1.1	3.9 ± 0.8	≤0.0001
t cholesterol (mmol/l)	4.53 ± 0.6	4.45 ± 0.6	ns
HDL (mmol/l)	1.41 ± 0.2	1.45 ± 0.2	ns
Triglycerides (mmol/l)	0.65 ± 0.1	0.71 ± 0.2	ns
Lipoprotein(a) (μmol/l)	0.09 ± 0.1	0.3 ± 0.2	<0.001
Adiponectin (mg/l)	16.3 ± 6	14.7 ± 5	ns
Leptin (μg/l)	52 ± 23	31 ± 19	≤0.01
Visfatin (μg/l)	13.3 ± 6	13 ± 5	ns

APTT, activated partial thromboplastin time; FGIR, fasting glucose-to-insulin ratio; ns, not significant ($P > 0.05$).

erozygous for the PT G20210A mutation ($P = 0.06$). One LGA but no AGA child was heterozygous for the Factor V Leiden (FVL) G1691A mutation. No child was homozygous for the prothrombin (PT) G20210A mutation or FVL polymorphism.

Correlation studies

On pooled data for LGA and AGA children, multiple regression analysis revealed negative correlation between birth weight *z* score and fibrinogen level ($t = -3.8$, $P < 0.01$), Lp(a) level ($t = -3.4$, $P < 0.01$), and IGFBP-3 level ($t = -2.5$, $P = 0.01$), and positive correlation between birth weight *z* score and I_F level ($t = 2.8$, $P = 0.01$) and HOMA-IR ($t = 2.9$, $P < 0.001$), independent of BMI or waist circumference *z* score.

Incidence of components of the MetS and other CVD risk factors

Of the LGA group, 9.7% fulfilled the criteria for MetS (≥3 components: waist circumference ≥90th percentile for age and sex for Greek children; BP ≥95th percentile for age, sex, and height; G_F >100 mg/dl; triglycerides >95th percentile; and HDL <5th percentile) (6,7), while no AGA child presented three components. In the AGA group, 54.5% of the children were completely free of components of MetS or risk factors for CVD (BP ≥90th percentile, I_F >15 μU/ml, fasting glucose-to-insulin ratio <7, HOMA-IR >2.83, or BMI >85th percentile) (6,8), while only 22.6% of the LGA children were free of components or risk factors ($P = 0.008$).

CONCLUSIONS — Children born LGA at term to nondiabetic, nonobese mothers are at significant risk of developing MetS. Diverse effects on CVD risk factors were observed in this group.

LGA children had significantly higher indexes of insulin resistance (I_F and HOMA-IR), independent of BMI or waist circumference *z* scores. The higher indexes of obesity, such as BMI and waist circumference found in this group may be attributed to earlier adiposity rebound (4,9).

Increase in fibrinogen level is associated with both chronic inflammation and insulin resistance in adults with MetS. LGA children had a significantly lower level of fibrinogen than AGA children. The negative association between fibrinogen level and birth weight suggests that excess intrauterine growth may significantly affect this factor in a direction different from the other factors contributing to CVD and MetS, and there is evidence that genetic factors may account for the negative association (10,11). A genetic influence may also be responsible for the lower levels of Lp(a) found in the LGA group.

The nature of the association found between LGA and mutation C677T in the MTHFR gene is unexplained and of unknown significance. This mutation is associated in the homozygous state with decreased specific MTHFR activity and elevation in homocysteine levels, which has been identified as an independent risk factor for myocardial infarction and mortality in patients with confirmed CVD (12). The significantly higher rate of homozygosity for the MTHFR-C677T mutation in the LGA group indicates a possible risk for hyperhomocysteinemia, which warrants further investigation. The independent negative relationship observed between IGFBP-3 and birth weight may imply a possible influence of excess intrauterine growth on IGFBP-3 level at prepuberty. Reduced plasma IGF-1 level is considered to reflect a higher risk for insulin resistance and CVD (13). In the present study, although LGA children had higher insulin resistance indexes, they also showed a trend toward higher IGF-1 levels. This may indicate a possible protective mechanism against development of insulin resistance (13).

In summary, diverse effects on CVD risk factors were observed in term LGA children at prepuberty. They had higher insulin resistance indexes and anthropometric obesity markers, but lower fibrinogen and Lp(a) levels than matched AGA children. They also had a higher prevalence of the MTHFR-C677T mutation. LGA offspring of nondiabetic, nonobese mothers warrant careful monitoring for evidence of MetS precursors throughout childhood and beyond.

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E.N.E. wrote the manuscript and researched the data. V.I.G. reviewed/edited the manuscript, contributed to the discussion, and researched the data. A.S.C. contributed to the discussion and researched the data. V.K.C., G.A.V., E.C.S., N.I.K., and E.T.B. researched the data. S.K.A. reviewed/edited the manuscript.

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