Pediatric RESEARCH

www.nature.com/pr

CLINICAL RESEARCH ARTICLE

Circulating growth-and-differentiation factor-15 in early life: relation to prenatal and postnatal growth and adiposity measurements

Marta Díaz[1,2], Laura Campderrós[1,3,4], Mariana P. Guimaraes[3], Abel López-Bermejo[5,6], Francis de Zegher[7,8], Francesc Villarroya[1,3,4] and Lourdes Ibáñez[1,2]

BACKGROUND: Growth-and-differentiation-factor-15 (GDF15) is a regulator of energy homeostasis. To determine the relationship between circulating GDF15 and parameters of metabolic health, we assessed longitudinally GDF15 concentrations in infants born either appropriate- (AGA) or small-for-gestational-age (SGA), the latter population known to be at risk for metabolic alterations, particularly after a rapid postnatal catch-up in weight.

METHODS: The study cohort consisted of 103 infants (70 AGA and 33 SGA). Assessments included body length, weight, and ponderal index (PI); fasting glucose, insulin, IGF-I, high-molecular-weight adiponectin, GDF15; and body composition (by absorptiometry) at birth, and at age 4, 12 and 24 months.

RESULTS: GDF15 levels at birth were significantly higher than those at each subsequent time point and were similar in AGA and SGA subjects. GDF15 concentrations dropped at age 4 months, more substantially in SGA infants, and continued to decline in both subgroups reaching adult concentrations by age 24 months. GDF15 levels correlated inversely with the changes in PI, IGF-I and body fat throughout follow-up.

CONCLUSIONS: Early life is associated with supra-adult concentrations of GDF15. The lower levels of GDF15 in SGA subjects may be an adaptive mechanism to promote catch-up in weight and might increase the risk for obesity later in life.

Pediatric Research (2020) 87:897–902; https://doi.org/10.1038/s41390-019-0633-z

INTRODUCTION

Growth-and-differentiation factor-15 (also known as macrophage inhibitory cytokine-C, MIC-1; or nonsteroidal anti-inflammatory drug-activated gene-1, NAG-1) is a member of the transforming growth factor superfamily β, and is secreted by the liver, kidney, and placenta, among other organs.[1] Although its biological functions are not completely understood, circulating GDF15 appears to operate as regulator of energy homeostasis, as a biomarker of pathological conditions, such as cancer, inflammatory disorders, cardiovascular disease and type 2 diabetes, and as a biomarker of mortality of any kind.[1–3] Indeed, GDF15 expression and circulating levels are increased in disorders associated with cell stress, where the upregulation of GDF15 would serve as a physiological counterregulatory mechanism.[4] Recently, a powerful anorexigenic role of GDF15 has been reported, and a specific GDF15 receptor, named GFRAL (glial-derived neurotrophic factor receptor alpha-like) has been identified in the brain as mediator of this effect.[5–7] The anorexigenic response elicited by high levels of GDF15 in cancer patients seems to account for appetite suppression within the anorexia/cachexia syndrome.[8]

Circulating GDF15 levels are several fold higher in healthy pregnant women as compared to nonpregnant women, possibly due to overexpression of GDF15 in placenta.[9] Although the physiological role of this adjustment is unknown, recent studies uncover an association between genetic variants within the GDF15 locus and hyperemesis gravidarum.[10] In pregnancies complicated with preeclampsia—a condition associated with augmented risks for cardiovascular events—maternal and cord blood GDF15 concentrations are further increased, suggesting GDF15 dysregulation.[11]

In accordance with the findings in adults, high levels of GDF15 have been described in severe pediatric diseases such as mitochondrial neuromyopathies and congenital heart disorders, where GDF15 has been postulated as a diagnostic biomarker.[12–14] Similarly, circulating GDF15 levels associate with markers of cardiometabolic risk in normal-weight children and in children with obesity.[15] However, despite the growing awareness of the importance of GDF15 in human pathophysiology, the ontogeny of GDF15 and its potential value as a biomarker of adiposity and/or metabolic health in early life has not been assessed so far. Here,

[1]Pediatric Research Institute Sant Joan de Déu, University of Barcelona, 08950 Esplugues, Barcelona, Spain; [2]Centro de Investigación Biomédica en Red de Diabetes y Enfermedades Metabólicas Asociadas (CIBERDEM), Health Institute Carlos III, Madrid, Spain; [3]Biochemistry and Molecular Biomedicine Department, Biomedicine Institute, University of Barcelona, 08028 Barcelona, Spain; [4]Network Biomedical Research Center of Physiopathology of Obesity and Nutrition (CIBEROBN), Health Institute Carlos III, 28029 Madrid, Spain; [5]Department of Pediatrics, Dr. Josep Trueta Hospital, 17007 Girona, Spain; [6]Girona Institute for Biomedical Research, Dr. Josep Trueta Hospital, 17007 Girona, Spain; [7]Pediatric & Adolescent Endocrinology, University Hospital Gasthuisberg, 3000 Leuven, Belgium and [8]Department of Development & Regeneration, University of Leuven, 3000 Leuven, Belgium

Correspondence: Lourdes Ibáñez (libanez@sjdhospitalbarcelona.org)

These authors contributed equally: Marta Díaz, Laura Campderrós

Received: 14 June 2019 Revised: 3 October 2019 Accepted: 13 October 2019
Published online: 23 October 2019

we explored whether circulating GDF15 levels relate to prenatal and early postnatal weight gain, and to parameters of metabolic health. To this aim, we assessed GDF15 concentrations from birth to age 24 months in a longitudinal cohort of apparently healthy infants born either appropriate- (AGA) or small-for-gestational-age (SGA) with spontaneous catch-up in growth, the latter population being known to exhibit increased risks for developing insulin resistance, central adiposity, and cardiovascular alterations, particularly when prenatal growth restraint is followed by rapid postnatal catch-up in weight.[16,17] The study design allowed to uncover for the first time the physiological variation in GDF15 levels in postnatal human development, and to determine their relationship with sex and birth weight, and with ponderal index (PI), endocrine-metabolic parameters and body composition throughout the first 2 years of life.

STUDY POPULATION AND METHODS

Study population

The study cohort consisted of 103 infants (70 AGA (49% girls) and 33 SGA (48% girls)) who were enrolled into two previous longitudinal, prospective studies (study 1 and study 2), with recruitment completed in September 2009 and in December 2014, respectively (Supplemental Fig. S1, flow chart). Study 1 assessed body composition and endocrine-metabolic variables of SGA infants, as compared with AGA-breastfed controls, in the first 2 years of life;[18] study 2 assessed DNA methylation in placenta and cord blood of SGA vs. AGA infants as well as endocrine-metabolic and body composition parameters also between birth and age 2 years.[19]

For the present substudy focusing on GDF15 assessment, the specific inclusion criteria were: (1) maternally uncomplicated, singleton pregnancy; (2) Caucasian origin; (3) birth at Hospital Sant Joan de Déu, Barcelona, at term (37–42 weeks); birth weight between 2.9 and 3.8 kg for AGA (range −1.1 and +1.1 SD), and between 1.9 and 2.6 kg for SGA (below −2 SD); (4) exclusive breastfeeding or formula-feeding in the first 4 months; (5) spontaneous catch-up in weight and length, defined as weight and length Z-score > −2.0 by age 1 year;[20] (6) cord serum and blood samples available at 4, 12 and 24 months for the measurement of GDF15; (7) written informed consent in Spanish/Catalan language, obtained in the third trimester of pregnancy. Exclusion criteria were: maternal hypertension, pre-eclampsia, gestational diabetes, alcohol or drug abuse, congenital malformations, syndromes and complications at birth (need for resuscitation, hypoglycemia, infections, or parenteral nutrition). The delivery rate by cesarean section was not significantly different in the AGA and SGA subgroups (19% and 27%, respectively ($p = 0.32$)); 13% of AGA infants and 45% of SGA infants were formula-fed during the first 4 months. The prevalence of maternal smoking during pregnancy was higher in the SGA than in the AGA subgroup (42 vs. 6%, respectively; $p = 0.0006$); however, fetal, uterine and placental doppler ultrasounds performed during the third trimester in these women showed no alterations.

Serum GDF15 was also measured cross-sectionally in 11 healthy AGA newborns (mean birth weight 3.3 kg) sampled at a mean age of 36 h, and in 18 pregnant women with singleton, uncomplicated pregnancies (mean gestational age, 35 weeks), who subsequently delivered healthy AGA infants (mean birth weight 3.3 kg). These women/newborns were included in a previous longitudinal study assessing circulating preadipocyte factor 1 in AGA and SGA infants.[21] In the 11 healthy AGA newborns, no longitudinal endocrine-metabolic and body composition assessments were performed. Moreover, serum GDF15 concentrations were also assessed cross-sectionally in a cohort of healthy adult volunteers ($n = 18$ (8 females), age, 41.4 ± 0.7 years; body mass index (BMI), 25.1 ± 0.5 kg/m^2 (mean ± SEM)).

Assessments

Gestational age was calculated according to the last menses and confirmed by first-trimester ultrasound (~10 weeks). Weight and length were measured at birth, and at 4, 12 and 24 months, and Z-scores derived by using the growth charts of Ferrández et al. suitable for the Spanish population.[19] Weight was measured with a beam balance (Seca, Hamburg, Germany) and length with a length board, the mean of three measurements was used for analysis.

Blood samples were obtained at birth from the umbilical cord (before placental separation), and in the $n = 103$ patients composing the longitudinal study (Supplemental Fig. S1), subsequent sampling at 4, 12 and 24 months was performed early in the morning after an overnight fast.

Serum glucose was measured by the glucose oxidase method. Lipids, insulin, and insulin-like growth factor-I (IGF-I) were assessed by immunochemiluminiscence (DPC IMMULITE 2500, Siemens, Germany); intra- and inter-assay coefficients of variation (CVs) were <10%;[16] the detection limit for IGF-I was 25 ng/mL. Insulin resistance was estimated with the homeostatic model assessment [fasting insulin (mU/L) × fasting glucose (mmol/L)/22.5]. High-molecular-weight (HMW) adiponectin was assessed by ELISA (R&D Systems, Minneapolis); intra- and inter-assay CVs being <9%. GDF15 levels were measured in duplicate using a specific human GDF15 ELISA kit (R&D Systems, Minneapolis), according to the manufacturer's instructions; intra- and inter-assay CVs were <6%.

Body composition was assessed at age 15 days and at 4, 12 and 24 months, by dual X-ray absorptiometry (DXA) with a Lunar Prodigy coupled to Lunar software (version 3.4/3.5; Lunar Corp, Madison, WI), adapted for infants; the assessments were performed during spontaneous sleep prior to feeding, and body fat, lean mass, and bone mineral content were determined.[18,19] CVs were <3% for fat and lean mass.

Statistics and ethics

Statistical analyses were performed with SPSS (version 23.0, IBM Corp, Armonk, NY). Results are expressed as mean ± SEM. The Kolmogorov−Smirnov test was applied to test for normal distribution. Differences between AGA and SGA subgroups were tested using unpaired t test or Mann−Whitney U test for normally distributed or nonparametric variables, respectively. Longitudinal changes in circulating GDF15 were examined by one-way analysis of variance (ANOVA) and Kruskal−Wallis post-hoc test. To assess simultaneously the effects of group and age on GDF15 levels, two-way ANOVA with post-hoc comparisons and Bonferroni correction was used. Correlation and step-wise multiple regression analyses were performed to study the associations between serum GDF15 and auxological, endocrine-metabolic and body composition variables. All metabolic parameters were log-transformed prior to performing the Pearson correlation analysis to ensure the normal distribution of the data. In multiple regression analyses, depending and predictor variables were standardized as Z-score. The effect sizes with 95% confidence intervals were added. Accepting and alpha risk of 0.05 and a beta risk of 0.20, the selected sample size allowed us to detect, in bilateral tests, significant differences in circulating GDF15 (between AGA and SGA subgroups) greater than or equal to 0.6 standard deviations, which are believed to be of clinical interest. Similarly, the sample size allowed us to detect, in bilateral tests, significant correlations between GDF15 and endocrine-metabolic and body composition parameters with a correlation coefficient of at least 0.28. The significance was set at $P < 0.05$.

The study was approved by the Institutional Review Board of Barcelona University, Hospital of Sant Joan de Déu; informed written consent was obtained from parents before delivery.

RESULTS

Table 1 shows the longitudinal data in the study population by birth weight subgroup. At birth, SGA newborns were lighter and

Table 1. Longitudinal data (0–24 months) in infants born appropriate-for-gestational-age (AGA, ($n = 70$)) or small-for-gestational age (SGA, ($N = 33$))

	At birth		4 months		Δ 0–4 months		12 months		Δ 0–12 months		24 months		Δ 0–24 months	
	AGA	SGA	AGA	SGA	AGA	SGA	AGA	SGA	AGA	SGA	AGA	SGA	AGA	SGA
Auxology														
Gestational age (wk)	**39.7 ± 0.1**	**38.8 ± 0.3**§	—	—	—	—	—	—	—	—	—	—	—	—
Weight (kg)	**3.3 ± .03**	**2.3 ± .04**‡	**6.7 ± 0.1**	**6.0 ± 0.1**§	**3.4 ± 0.1**	**3.7 ± 0.1***	**9.8 ± 0.2**	**8.8 ± 0.2**‡	6.5 ± 0.2	6.5 ± 0.2	**12.1 ± 0.2**	**11.3 ± 0.2**§	8.8 ± 0.2	8.9 ± 0.2
Weight SDS	**0.01 ± 0.1**	**−2.3 ± 0.1**‡	**−0.3 ± 0.2**	**−1.2 ± 0.2**§	**−0.4 ± 0.2**	**1.1 ± 0.2**‡	**−0.03 ± 0.2**	**−1.1 ± 0.1**‡	**−0.05 ± 0.2**	**1.2 ± 0.1**‡	**−0.2 ± 0.1**	**−0.9 ± 0.2**§	**−0.3 ± 0.1**	**1.5 ± 0.1**‡
Length (cm)	**50.0 ± 0.2**	**46.2 ± 0.3**‡	**62.9 ± 0.5**	**60.8 ± 0.6***	**12.9 ± 0.6**	**14.6 ± 0.6**‡	75.2 ± 0.5	74.3 ± 0.7	**25.2 ± 0.5**	**28.1 ± 0.8**§	85.3 ± 0.7	84.5 ± 0.9	**35.3 ± 0.8**	**38.2 ± 0.9***
Length SDS	**0.1 ± 0.1**	**−1.7 ± 0.2**‡	**−0.3 ± 0.2**	**−1.2 ± 0.2***	**−0.3 ± 0.3**	**0.4 ± 0.3**§	0.2 ± 0.2	−0.2 ± 0.3	**0.1 ± 0.2**	**1.5 ± 0.4**§	−0.6 ± 0.2	−0.9 ± 0.3	**−0.7 ± 0.3**	**0.8 ± 0.3**‡
PI (kg/m^3)	**26.3 ± 0.3**	**23.6 ± 0.5**‡	27.1 ± 0.5	26.9 ± 0.6	**0.5 ± 0.6**	**3.3 ± 0.7**§	**23.1 ± 0.4**	**21.7 ± 0.6**§	−3.6 ± 0.5	−1.9 ± 0.7	**19.8 ± 0.4**	**18.3 ± 0.4**§	**−6.8 ± 0.5**	**−4.6 ± 0.9***
Endocrine-metabolic variables														
Glucose (mmol/L)	4.6 ± 0.1	4.7 ± 0.3	4.9 ± 0.1	4.9 ± 0.1	0.4 ± 0.1	0.2 ± 0.3	4.6 ± 0.1	4.6 ± 0.1	0.01 ± 0.1	−0.1 ± 0.4	4.5 ± 0.1	4.5 ± 0.1	0.1 ± 0.2	−0.2 ± 0.3
Insulin (pmol/L)	**40.9 ± 4.9**	**26.4 ± 6.2**§	50.0 ± 7.6	46.5 ± 7.6	9.0 ± 1.4	20.1 ± 11.1	29.2 ± 5.5	22.9 ± 5.5	−11.8 ± 6.9	−3.5 ± 9.0	**22.9 ± 3.5**	**11.8 ± 2.8***	−18.0 ± 6.2	−12.5 ± 6.2
HOMA-IR	**1.2 ± 0.2**	**0.7 ± 0.2**§	1.7 ± 0.3	1.5 ± 0.3	0.5 ± .03	0.4 ± 0.5	0.9 ± 0.2	0.7 ± 0.2	−0.3 ± 0.3	−0.4 ± 0.4	**0.7 ± 0.1**	**0.3 ± 0.1***	−0.5 ± 0.2	−0.8 ± 0.4
Triglycerides (mmol/L)	—	—	**1.7 ± 0.1**	**1.2 ± 0.1***	—	—	1.2 ± 0.1	0.9 ± 0.1	—	—	0.8 ± 0.1	0.9 ± 0.1	—	—
LDL-C (mmol/L)	—	—	2.0 ± 0.1	1.8 ± 0.1	—	—	2.5 ± 0.1	2.2 ± 0.2	—	—	2.5 ± 0.1	2.2 ± 0.1	—	—
HDL-C (mmol/L)	—	—	1.0 ± 0.0	1.1 ± 0.1	—	—	0.9 ± 0.0	1.1 ± 0.1	—	—	1.1 ± 0.1	1.0 ± 0.1	—	—
IGF-I (nmol/L)	**7.4 ± 0.4**	**5.0 ± 0.4**‡	**5.2 ± 0.3**	**8.4 ± 1.3**§	**−2.2 ± 0.1**	**3.4 ± 1.2**‡	7.7 ± 0.5	8.1 ± 0.8	**0.3 ± 0.6**	**3.3 ± 0.6**§	8.9 ± 0.5	9.3 ± 0.9	**1.3 ± 0.6**	**4.3 ± 0.6***
HMW adiponectin (mg/L)	29.9 ± 1.3	29.4 ± 2.6	**27.3 ± 1.9**	**36.9 ± 3.8***	**−2.6 ± 0.4**	**7.5 ± 3.6**§	17.6 ± 1.1	19.9 ± 2.4	−12.3 ± 1.8	−9.6 ± 3.2	17.0 ± 1.3	19.4 ± 2.2	−12.9 ± 2.0	−10.5 ± 4.5
GDF15 (pg/mL)	3095 ± 191	3183 ± 164	**555 ± 25**	**469 ± 34**§	−2540 ± 209	−2714 ± 156	441 ± 16	426 ± 25	−2654 ± 200	−2757 ± 155	394 ± 15	357 ± 14	−2701 ± 196	−2826 ± 164
Body composition (DXA)														
Age at DXA (days)	15 ± 1	15 ± 1	131 ± 2	136 ± 3	116 ± 1	121 ± 3	391 ± 6	409 ± 7	376 ± 6	394 ± 7	748 ± 4	759 ± 25	733 ± 4	744 ± 25
Fat mass (kg)	**0.8 ± .03**	**0.5 ± .02**‡	2.5 ± 0.1	2.4 ± 0.1	1.8 ± 0.1	1.9 ± 0.1	3.4 ± 0.1	3.2 ± 0.1	2.6 ± 0.1	2.7 ± 0.1	**3.9 ± 0.1**	**3.2 ± 0.2**§	3.1 ± 0.1	2.7 ± 0.1
Abdominal fat (kg)	**0.04 ± .002**	**0.02 ± .002**‡	0.14 ± 0.01	0.14 ± 0.08	0.11 ± 0.01	0.12 ± 0.01	0.18 ± .01	0.20 ± .04	0.14 ± .01	0.18 ± .04	**0.19 ± .01**	**0.15 ± .01***	0.15 ± .01	0.13 ± .01
Lean mass (kg)	**3.1 ± .04**	**2.3 ± .05**‡	**4.4 ± 0.1**	**3.9 ± 0.1**§	1.3 ± 0.1	1.6 ± 0.1	**6.7 ± 0.1**	**5.7 ± 0.3**‡	3.6 ± 0.1	3.4 ± 0.3	**8.5 ± 0.2**	**7.8 ± 0.2***	5.4 ± 0.2	5.5 ± 0.2

PI ponderal index, *HOMA-IR* homeostasis model assessment—insulin resistance, *LDL-C* LDL-cholesterol, *HDL-C* HDL-cholesterol, *IGF-I* insulin-like growth factor-I, *HMW adiponectin* high-molecular-weight adiponectin, *GDF15* growth differentiation factor 15
Values are mean ± SEM. *$P < 0.05$, $^{\S}P < 0.01$, $^{\ddagger}P < 0.001$ vs. AGA
The bold values highlight the statistical differences

shorter, displayed lower levels of insulin and IGF-I in cord blood, and less fat and lean mass, confirming previous reports.[18,22] The catch-up in fat mass at age 4 months in SGA infants was accompanied by higher levels of IGF-I and HMW-adiponectin, as compared to AGA infants; both parameters subsequently normalized by age 12 months. At age 24 months, SGA infants remained lighter, displayed less fat mass and less lean mass and lower insulin concentrations; HMW-adiponectin concentrations were significantly higher in formula-fed than in breastfed SGA infants, who were also less abdominally adipose (Supplementary Table S1).

Figure 1 depicts the longitudinal outcome of GDF15 concentrations. Circulating GDF15 levels were not influenced by sex, type of delivery (data not shown), or early nutrition (breast- or formula-feeding in the first 4 months of life (Supplementary Table S1)), and thus the results within each study subgroup were pooled. GDF15 levels at birth were significantly higher than those at each subsequent time point ($P < 0.001$), were similar in AGA and SGA infants, and were lower than those measured in maternal blood in late gestation (12,643 ± 154 pg/mL; $P < 0.05$ vs. AGA and SGA).

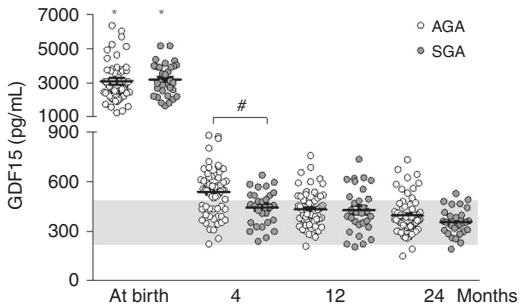

Fig. 1 GDF15 longitudinal data in the study population (70 AGA and 33 SGA). The upper and lower limits of the gray zone correspond to a *Z*-score of +1 and −1, respectively, in healthy adults ($n = 18$; age, 41.4 ± 0.7 years; BMI, 25.1 ± 0.5 kg/m^2; circulating GDF15, 355 ± 18 pg/mL (mean ± SEM)). *$P < 0.001$ between values at birth and those at 4, 12 and 24 months; #$P < 0.01$ for AGA vs. SGA

GFD15 levels were still high at a mean age of 36 h (2861 ± 319 pg/mL), dropped by age 4 months, and experienced afterwards a progressive decrease reaching adult levels by the age of 24 months. The fall in serum GDF15 from 4 to 24 months was significant in the AGA and SGA subgroups ($P < 0.0001$ and $P = 0.007$, respectively). At each time point, GDF15 concentrations tended to be lower in SGA infants; these differences were particularly evident at age 4 months, reaching statistical significance ($P = 0.008$ vs. AGA). Longitudinal GDF15 changes between 4 and 24 months were significantly influenced by both birth weight group ($P = 0.01$) and age ($P < 0.0001$), and explained 12% of GDF15 variability. However, when cord GDF15 levels were added in the analysis, the effect of birth weight was lost and only age remained significant ($P < 0.0001$).

Table 2 shows the correlations between circulating GDF15 and clinical, biochemical and body composition variables throughout follow-up. At birth, GDF15 levels correlated negatively with PI and with IGF-I concentrations; at age 12 months, circulating GDF15 correlated inversely with PI, and positively with HMW-adiponectin; at age 24 months, GDF15 associated positively with HDL-cholesterol and negatively with PI, insulin, HOMA-IR, IGF-I and triglycerides. Circulating GDF15 levels correlated inversely with the 0–4 months, 0–12 months and 0–24 months changes in PI and IGF-I, the strongest correlation being with the 0–4-month change in PI ($r = -0.463$; $p = 0.0001$). In addition, serum GDF15 levels associated negatively with the 0–24-month change in total fat and abdominal fat.

Serum GDF15 at 4 months, and total fat and IGF-I levels at age 12 months, independently predicted fat mass at age 24 months in multiple regression analysis, together explaining 64% of its variance (Table 3). In addition, the percentage of total fat at age 24 months was independently explained by PI at birth, GDF15 at 4 months and by IGF-I and total fat at age 12 months.

DISCUSSION

To our knowledge, this is the first study assessing longitudinal changes in circulating GDF15 concentrations throughout the first 2 years of life, in apparently healthy infants born AGA or SGA from

Table 2. Correlations of circulating GDF15 concentrations with endocrine-metabolic, and imaging parameters, at birth and throughout follow-up to age 24 months in children born appropriate-for-gestational-age (AGA, $n = 70$) or small-for-gestational-age (SGA, $n = 33$)

	At birth		4 months		Δ 0–4 months		12 months		Δ 0–12 months		24 months		Δ 0–24 months	
	r	*P*	*r*	*P*	*r*	*P*	*r*	*P*	*r*	*P*	*r*	*P*	*r*	*P*
Weight	−0.134	0.183	−0.045	0.669	−0.139	0.195	0.048	0.649	−0.042	0.697	0.103	0.334	−0.144	0.193
Length	−0.119	0.249	−0.103	0.330	0.064	0.554	0.096	0.363	0.112	0.304	0.177	0.097	0.065	0.562
PI	−0.157	0.124	0.055	0.597	**−0.383**	**0.0004**	**−0.231**	**0.028**	**−0.249**	**0.019**	**−0.252**	**0.019**	**−0.325**	**0.002**
Fat mass	−0.093	0.359	−0.083	0.426	−0.196	0.067	−0.072	0.499	−0.069	0.518	0.119	0.268	**−0.303**	**0.007**
Abd fat mass	−0.084	0.408	0.030	0.775	−0.171	0.119	−0.068	0.520	−0.087	0.423	−0.031	0.776	**−0.297**	**0.009**
Lean mass	−0.121	0.234	0.063	0.545	−0.143	0.189	−0.154	0.146	−0.144	0.183	0.012	0.910	−0.106	0.335
Insulin	0.102	0.319	0.154	0.148	0.011	0.922	−0.040	0.703	0.182	0.097	**−0.293**	**0.006**	0.013	0.905
HOMA-IR	0.102	0.373	0.151	0.156	−0.078	0.528	−0.031	0.769	0.136	0.258	**−0.276**	**0.009**	0.099	0.424
IGF-I	**−0.455**	**0.0001**	−0.127	0.230	**−0.421**	**0.0001**	−0.074	0.482	**−0.357**	**0.0007**	**−0.323**	**0.002**	**−0.370**	**0.0005**
HMW adipo	0.067	0.505	0.161	0.252	−0.146	0.380	**0.289**	**0.004**	−0.028	0.796	0.112	0.402	0.064	0.631
LDL-cholesterol	—	—	—	0.682	—	—	—	0.344	—	—	0.063	0.639	—	—
HDL-cholesterol	—	—	—	0.469	—	—	—	0.340	—	—	**0.355**	**0.009**	—	—
Triglycerides	—	—	—	0.199	—	—	—	0.269	—	—	**−0.353**	**0.009**	—	—

PI ponderal index, *Abd fat mass* abdominal fat mass, *HOMA-IR* homeostasis model assessment—insulin resistance, *IGF-I* insulin-like growth factor-I, *HMW adipo* high-molecular-weight adiponectin
All metabolic parameters were log-transformed prior to correlation analysis
The bold values highlight the statistical differences

Table 3. Multivariate linear model of total fat (a) and % total fat (b) at age 24 months and selected endocrine-metabolic parameters

	(a) Total fat *Z*-score at age 24 months				(b) % Total fat *Z*-score at age 24 months			
	Beta	Sig.	R^2	95% CI	Beta	Sig.	R^2	95% CI
GDF15 *Z*-score at 4 months	−0.316	0.008	0.096	−0.51 to −0.09	−0.316	0.002	0.105	−0.51 to −0.08
Total fat *Z*-score at 12 months	0.652	<0.001	0.412	0.51−0.76	0.483	<0.001	0.168	0.32−0.63
IGF-I *Z*-score at 12 months	0.338	0.001	0.132	0.14−0.51	0.250	0.016	0.046	0.03−0.42
Ponderal index *Z*-score at birth	—	—	—	—	0.236	0.020	0.054	0.05−0.44

(a) Nonpredictive variables: Ponderal index (PI) at 0, 4 or 12 months, total fat at 0 or 4 months, abdominal fat at 0, 4 or 12 months, lean mass at 0, 4 or 12 months, GDF15 at 0 or 12 months, HOMA-IR and HMW-adipo at 0, 4 or 12 months and IGF-I at 0 or 4 months
(b) Nonpredictive variables: Ponderal index (PI) at 4 or 12 months, total fat at 0 or 4 months, abdominal fat at 0, 4 or 12 months, lean mass at 0, 4 or 12 months, GDF15 at 0 or 12 months, HOMA-IR and HMW-adipo at 0, 4 or 12 months and IGF-I at 0 or 4 months

uncomplicated, term pregnancies. Here, we report that the high GDF15 concentrations at birth drop at age 4 months, being this decline more pronounced in SGA infants, and that adult GDF15 levels are reached by age 24 months. Moreover, GDF15 levels in the first and second year of life associate inversely with the changes in markers of postnatal catch-up.

Cord GDF15 levels were comparable in AGA and SGA newborns, represented only 25% of those found in maternal blood in late gestation,[23] and were about tenfold higher than in our cohort of adults, who in addition, exhibited circulating GDF15 concentrations similar to those previously described in other populations.[24,25] Cord GDF15 concentrations were in the range of those reported in small groups of newborns with unspecified birth weight or gestational age,[11] and decreased slightly thereafter. Although the placenta secretes large amounts of GDF15,[26] the maintenance of high GDF15 levels at a mean age of 36 h of postnatal life, and the reported short half-life of GDF15[27] suggest a neonatal rather than placental origin of GDF15 in newborns.

At age 4 months, GDF15 concentrations had dropped in AGA and SGA infants; being this fall significantly more pronounced in SGA subjects. Those differences might be a developmental programmed event that could contribute in part to postnatal catch-up in weight in SGA individuals.

GDF15 is increasingly recognized as a key circulating regulatory factor of long-term energy homeostasis and inflammation in adults,[28–30] and recent findings—albeit not universally accepted—tend to support the anorexigenic effect of this cytokine through central actions.[1,24] The relatively lower levels of GDF15 in SGA infants at age 4 months would be consistent with a decrease of the inhibitory action exerted by this cytokine on hypothalamic feeding settings and on energy expenditure during the period where catch-up is taking place[31] and thus, when a parallel increase in caloric intake is expected. Consequently, it can be speculated that the reduced GDF15 levels in SGA infants early in postnatal life may impair central mechanisms of satiety and thereby contribute to the long-term promotion of a positive energy balance and thus, potentially, to the increased risks for obesity and diabetes. Long-term follow-up of this population will disclose whether they subsequently experience GDF15 upregulation, a mechanism that appears to counteract progressive weight gain in experimental models.[32]

The catch-up in weight in SGA infants between birth and age 4 months was accompanied by increasing levels of IGF-I and HMW-adiponectin, all these features having been previously reported in this population, particularly when SGA infants are formula-fed.[18] Indeed, HMW-adiponectin was higher in formula-fed than in breastfed SGA infants, who were also less abdominally adipose. The higher levels of HMW-adiponectin may—at least in part—drive the catch-up of fat, because this adipokine can enhance early lipogenesis.[33] Circulating GDF15 concentrations throughout follow-up correlated negatively with the changes in IGF-I, a marker of anabolism.[34] The positive association between circulating GDF15 and HMW-adiponectin at age 12 months and the negative correlation of GDF15 with markers of insulin resistance at age 24 months further support the relationship between low levels of GDF15 in childhood and the development of an adverse metabolic status. This prospect is strengthened by the finding of a predictive value of early circulating GDF15 levels (at age 4 months) on body adiposity (at age 24 months).

In pediatric populations, abnormally high GDF15 concentrations have been reported in association with severe pathological conditions such as neuromuscular disorders of mitochondrial origin,[12] pulmonary hypertension secondary to congenital heart disease,[13] and sickle cell disease.[35] However, in apparently healthy subjects with prenatal growth restraint who develop spontaneous postnatal catch-up, the lower GDF15 levels should be viewed with concern, given its possible association with the future development of obesity, hepato-visceral fat excess, and metabolic disorders. Along these lines, it has been recently shown that endogenous administration of GDF15 protects against the progression of steatohepatitis,[36] and increases the expression of thermogenic and lipolytic genes in both brown and white adipose tissue in animal models.[37] Furthermore, GDF15 deficiency drives obesity, hepatic lipid deposition, and associated comorbidities.[38,39]

The limitations of the present study include the relatively small size of the studied population, the absence of follow-up beyond age 2 years, and the lack of a non-catch-up SGA subgroup for comparisons. The strengths include being the first assessment of GDF15 levels in early life, the longitudinal design, the homogeneity within the two study subgroups, avoiding the bias introduced by birth weight overlapping between subpopulations, and the use of the same methodology over time.

In summary, early life is associated with higher levels of GDF15 relative to adulthood. The relatively reduced GDF15 levels in early postnatal life in infants born SGA may represent an adaptive mechanism to promote food intake and thus, postnatal catch-up in weight. Further research will be needed to determine whether such adaptation may exert long-term programming effects favoring a sustained positive energy balance, and may thus potentially increase the subsequent risks for obesity and/or diabetes.

ACKNOWLEDGEMENTS

M.D. and L.I. are investigators of CIBERDEM (www.ciberdem.org); F.V. and L.C. are investigators of CIBEROBN (www.ciberobn.org) (National Health Institute Carlos III, Madrid, Spain). A.L.-B. is an investigator of the 13 Fund for Scientific research (Ministry of Education and Science, Spain). M.P.G. is funded by the São Paulo Research Fundation (FAPESP), Brazil (Grant 2017/26509-8). F.dZ. is a clinical investigator from the Clinical Research Council of Leuven University Hospitals, Belgium. This study was supported by grants from the "Instituto de Salud Carlos III" and the "Fondo Europeo de Desarrollo Regional" (FEDER), Madrid, Spain (PI08/0443, PI11/02403, and PI18/00109), and from Ministerio de Ciencia, Innovación y Universidades (SAF2017-85722-R).

AUTHOR CONTRIBUTIONS

M.D. and L.C. researched and analyzed data, contributed to data interpretation and wrote the manuscript. M.P.G. contributed to the GDF15 analysis. A.L.-B. reviewed/edited the manuscript. F.dZ. reviewed/edited the manuscript. F.V. contributed to study design, and wrote/reviewed/edited the manuscript. L.I. contributed to study design, and wrote/reviewed/edited the manuscript. Each author listed in the manuscript has seen and approved the submission of this version of the manuscript and takes full responsibility for the contents.

ADDITIONAL INFORMATION

The online version of this article (https://doi.org/10.1038/s41390-019-0633-z) contains supplementary material, which is available to authorized users.

Competing interests: The authors declare no competing interests.

Publisher's note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.

REFERENCES

1. Tsai, V. W. W. et al. The MIC-1/GDF15-GFRAL pathway in energy homeostasis: implications for obesity, cachexia, and other Associated diseases. *Cell Metab.* **28**, 353–368 (2018).
2. Adela, R. & Banerjee, S. K. GDF-15 as a target and biomarker for diabetes and cardiovascular diseases: a translational prospective. *J. Diabetes Res.* **2015**, 490842 (2015).
3. Hagström, E. et al. Growth Differentiation Factor 15 predicts all-cause morbidity and mortality in stable coronary heart disease. *Clin. Chem.* **63**, 325–333 (2017).
4. Carstensen, M. et al. Macrophage inhibitory cytokine-1 is increased in individuals before type 2 diabetes diagnosis but is not an independent predictor of type 2 diabetes: the Whitehall II study. *Eur. J. Endocrinol.* **162**, 913–917 (2010).
5. Hsu, J. Y. et al. Non-homeostatic body weight regulation through a brainstem-restricted receptor for GDF15. *Nature* **550**, 255–259 (2017).
6. Yang, L. et al. GFRAL is the receptor for GDF15 and is required for the anti-obesity effects of the ligand. *Nat. Med.* **23**, 1158–1166 (2017).
7. Mullican, S. E. et al. GFRAL is the receptor for GDF15 and the ligand promotes weight loss in mice and nonhuman primates. *Nat. Med.* **23**, 1150–1157 (2017).
8. Tsai, V. W. et al. Anorexia-cachexia and obesity treatment may be two sides of the same coin: role of the TGF-b superfamily cytokine MIC-1/GDF15. *Int. J. Obes. (Lond.).* **40**, 193–197 (2016).
9. Moore, A. G. et al. The transforming growth factor-ss superfamily cytokine macrophage inhibitory cytokine-1 is present in high concentrations in the serum of pregnant women. *J. Clin. Endocrinol. Metab.* **85**, 4781–4788 (2000).
10. Fejzo, M. S. et al. Placenta and appetite genes GDF15 and IGFBP7 are associated with hyperemesis gravidarum. *Nat. Commun.* **9**, 1178 (2018).
11. Sugulle, M. et al. Circulating and placental growth-differentiation factor 15 in preeclampsia and in pregnancy complicated by diabetes mellitus. *Hypertension* **54**, 106–112 (2009).
12. Montero, R. et al. GDF-15 is elevated in children with mitochondrial diseases and is induced by mitochondrial dysfunction. *PLoS ONE* **11**, e0148709 (2016).
13. Li, G. et al. Plasma Growth Differentiation Factor-15 is a potential biomarker for pediatric pulmonary arterial hypertension associated with congenital heart disease. *Pediatr. Cardiol.* **38**, 1620–1626 (2017).
14. Wang, T. et al. GDF15 is a heart-derived hormone that regulates body growth. *EMBO Mol. Med.* **9**, 1150–1164 (2017).
15. Yuca, S. A. et al. The relationship between metabolic parameters, cardiac parameters and MIC-1/GDF15 in obese children. *Exp. Clin. Endocrinol. Diabetes* **125**, 86–90 (2017).
16. Sebastiani, G. et al. The sequence of prenatal growth restraint and postnatal catch-up growth leads to a thicker intima-media and more pre-peritoneal and hepatic fat by age 3–6 years. *Pediatr. Obes.* **1**, 251–257 (2016).
17. Sebastiani, G. et al. The sequence of prenatal growth restraint and postnatal catch-up growth: normal heart but thicker intima-media and more pre-peritoneal fat in late infancy. *Pediatr. Obes.* **14**, e12476e (2018).
18. de Zegher, F. et al. Body composition and circulating high-molecular-weight adiponectin and IGF-I in infants born small for gestational age: breast- versus formula-feeding. *Diabetes* **61**, 1969–1973 (2012).
19. Díaz, M. et al. Placental and cord blood methylation of genes envolved in energy homeostasis: association with fetal growth and neonatal body composition. *Diabetes* **66**, 779–784 (2017).
20. Clayton, P. E. et al. Management of the child born small for gestational age through to adulthood: a consensus statement of the International Societies of Pediatric Endocrinology and the Growth Hormone Research Society. *J. Clin. Endocrinol. Metab.* **92**, 804–810 (2007).
21. de Zegher, F. et al. Abundance of circulating preadipocyte factor-1 in early life. *Diabetes Care* **35**, 848–849 (2012).
22. Díaz, M. et al. GLP-1 and IGF-I levels are elevated in late infancy in low birth weight infants, independently of GLP-1 receptor polymorphisms and neonatal nutrition. *Int J. Obes. (Lond.).* **42**, 915–918 (2018).
23. Kaitu'u-Lino, T. J. et al. Plasma MIC-1 and PAPP-a levels are decreased among women presenting to an early pregnancy assessment unit, have fetal viability confirmed but later miscarry. *PLoS ONE* **8**, e72437 (2013).
24. Tsai, V. W. et al. Serum levels of human MIC-1/GDF15 vary in a diurnal pattern, do not display a profile suggestive of a satiety factor and are related to BMI. *PLoS ONE* **10**, e0133362 (2015).
25. Wollert, K. C., Kempf, T. & Wallentin, L. Growth Differentiation Factor 15 as a biomarker in cardiovascular disease. *Clin. Chem.* **63**, 140–151 (2017).
26. Segerer, S. E. et al. MIC-1 (a multifunctional modulator of dendritic cell phenotype and function) is produced by decidual stromal cells and trophoblasts. *Hum. Reprod.* **27**, 200–209 (2012).
27. Xiong, Y. et al. Long-acting MIC-1/GDF15 molecules to treat obesity: evidence from mice to monkeys. *Sci. Transl. Med* **9**, eaan8732 (2017). pii.
28. Tsai, V. W. et al. TGF-b superfamily cytokine MIC-1/GDF15 is a physiological appetite and body weight regulator. *PLoS ONE* **8**, e55174 (2013).
29. Chung, H. K. et al. Growth differentiation factor 15 is a myomitokine governing systemic energy homeostasis. *J. Cell Biol.* **216**, 149–165 (2017).
30. Mullican, S. E. & Rangwala, S. M. Uniting GDF15 and GFRAL: therapeutic opportunities in obesity and beyond. *Trends Endocrinol. Metab.* **29**, 560–570 (2018).
31. Hokken-Koelega, A. C. et al. Children born small for gestational age: do they catch up? *Pediatr. Res.* **38**, 267–271 (1995).
32. Patel, S. et al. GDF15 provides an endocrine signal of nutritional stress in mice and humans. *Cell Metab.* **29**, 707–718 (2019).
33. Körner, A. et al. New predictors of the metabolic syndrome in children-role of adipocytokines. *Pediatr. Res* **61**, 640–645 (2007).
34. Rosenfeld, R. G. & Hwa, V. The growth hormone cascade and its role in mammalian growth. *Horm. Res.* **1**(Suppl 2), 36–40 (2009).
35. Tantawy, A. A. et al. Growth differentiation factor-15 in young sickle cell disease patients: relation to hemolysis, iron overload and vascular complications. *Blood Cells Mol. Dis.* **53**, 189–193 (2014).
36. Kim, K. H. et al. Growth differentiation factor 15 ameliorates nonalcoholic steatohepatitis and related metabolic disorders in mice. *Sci. Rep.* **8**, 6789 (2018).
37. Chrysovergis, K. et al. NAG-1/GDF15 prevents obesity by increasing thermogenesis, lipolysis and oxidative metabolism. *Int J. Obes. (Lond.)* **38**, 1555–1564 (2014).
38. Tran, T. et al. GDF15 deficiency promotes high fat diet-induced obesity in mice. *PLoS ONE* **13**, e0201584 (2018).
39. Tsai, V. W., et al. GDF15 mediates adiposity resistance through actions of GFRAL neurons in the hindbrain AP/NTS. *Int. J. Obes.* (*Lond.*). https://doi.org/10.1038/s41366-019-0365-5 (2019)