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ARTICLE

Can placental growth factors explain birthweight variation in offspring of women with type 1 diabetes?

Siobhan Bacon[1,2] · Dylan Burger[3] · Mayur Tailor[3] · J. Johanna Sanchez[4] · George Tomlinson[2,5] · Helen R. Murphy[6,7,8] · Denice S. Feig[1,2,9] · on behalf of the CONCEPTT Collaborative Group

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Abstract

Aims/hypothesis Maternal hyperglycaemia alone does not explain the incidence of large offspring amongst women with type 1 diabetes. The objective of the study was to determine if there is an association between placental function, as measured by angiogenic factors, and offspring birthweight *z* score in women with type 1 diabetes.

Methods This cohort study included samples from 157 Continuous Glucose Monitoring in Pregnant Women with Type 1 Diabetes (CONCEPTT) trial participants. Correlations were estimated between birthweight *z* score and placental growth factor (PlGF) and soluble fms-like tyrosine kinase (sFlt-1) levels measured at baseline and at 24 and 34 weeks of gestation. Linear regression was used to assess the relationship between birthweight *z* score and placental health, as measured by PlGF and sFlt-1/PlGF ratio, stratified by glycaemic status (continuous glucose monitoring and HbA_{1c} measures) and adjusted for potential confounders of maternal BMI, smoking and weight gain. Higher PlGF levels and lower sFlt-1/PlGF ratios represent healthy placentas, while lower PlGF levels and higher sFlt-1/PlGF ratios represent unhealthy placentas.

Results Among CONCEPTT participants, the slopes relating PlGF levels to birthweight *z* scores differed according to maternal glycaemia at 34 weeks of gestation ($p = 0.003$). With optimal maternal glycaemia ($HbA_{1c} < 48$ mmol/mol [6.5%]/ or continuous glucose monitoring time above range $\leq 30\%$), birthweight *z* scores were reduced towards zero (normal weight) with increasing PlGF values (representing a healthy placenta), and increased with decreasing PlGF values. With suboptimal glycaemic status ($HbA_{1c} \geq 48$ mmol/mol [6.5%] or time above range $> 30\%$), increasing PlGF values were associated with heavier infants. Those with a healthy placenta (PlGF > 100) and suboptimal glycaemic control had a higher mean *z* score (2.45) than those with an unhealthy placenta (mean *z* score = 1.86). Similar relationships were seen when using sFlt-1/PlGF ratio as a marker for a healthy vs unhealthy placenta.

Conclusions/interpretation In women with type 1 diabetes, infant birthweight is influenced by both glycaemic status and placental function. In women with suboptimal glycaemia, infant birthweight was heavier when placentas were healthy. Suboptimal placental function should be considered in the setting of suboptimal glycaemia and apparently 'normal' birthweight.

Keywords Diabetes · Diabetes mellitus · Placenta growth factor · Pregnancy · Pregnancy in diabetics · Pregnancy outcomes · Type 1 diabetes · Vascular endothelial growth factor receptor 1

✉ Denice S. Feig
d.feig@utoronto.ca

[1] Mount Sinai Hospital, Sinai Health System, Toronto, ON, Canada

[2] Department of Medicine, University of Toronto, Toronto, ON, Canada

[3] Department of Cellular and Molecular Medicine, University of Ottawa, Ottawa, ON, Canada

[4] Sunnybrook Research Institute, Toronto, ON, Canada

[5] University Health Network, Toronto, ON, Canada

[6] Cambridge University Hospitals NHS Foundation Trust, Cambridge, UK

[7] Norwich Medical School, University of East Anglia, Norwich, UK

[8] Women's Health Academic Centre, Division of Women and Children's Health, Kings College London, London, UK

[9] Lunenfeld-Tanenbaum Research Institute, Toronto, ON, Canada

Research in context

What is already known about this subject?

- Maternal glycaemia alone does not explain birthweight variation amongst infants of women with type 1 diabetes
- The fetoplacental circulation system impacts on fetal size and is influenced by placental angiogenic factors (PlGF and sFlt-1), with high PlGF or a low sFlt-1/PlGF ratio representing a healthy placenta and low PlGF or a high sFlt-1/PlGF ratio representing an unhealthy placenta

What is the key question?

- Do alterations in placental angiogenic factors (PlGF and sFlt-1) contribute to variations in infant birthweight amongst women with type 1 diabetes?

What are the new findings?

- Infant birthweight was variably affected by placental angiogenic factors, depending on maternal glycaemia
- Infants of women with suboptimal glycaemia were heavier when placentas were healthy (high PlGF or a low sFlt-1/PlGF ratio), independent of maternal BMI, smoking and weight gain
- Infants of women with suboptimal glycaemia were lighter when placentas were less healthy (low PlGF or a high sFlt-1/PlGF ratio)

How might this impact on clinical practice in the foreseeable future?

- Additional obstetric surveillance may be applicable in women with HbA_{1c} ≥48 mmol/mol (6.5%) or >30% time above range (7.8 mmol/l [140 mg/dl]) and apparently 'normal' fetal growth, as this may infer suboptimal placental function. More frequent obstetric surveillance (for example, utilising Doppler artery studies) in this group may be appropriate

Abbreviations

CGM	Continuous glucose monitoring
CONCEPTT	Continuous Glucose Monitoring in Pregnant Women with Type 1 Diabetes trial
GROW	Gestation-related optimal weight
LGA	Large for gestational age
NICU	Neonatal intensive care unit
NPV	Negative predictive value
PlGF	Placental growth factor
sFlt-1	Soluble fms-like tyrosine kinase
SGA	Small for gestational age
TAR	Time above range
% TAR	Percentage of time spent above the range
TIR	Time in range
%TIR	Percentage of time spent in range
VEGF	Vascular endothelial growth factor

Introduction

The determinants of fetal growth include genetic make-up and maternal nutrient availability, as well as the capacity of the placenta to adequately transfer both oxygen and nutrients to the fetus and their endocrine modulation [1]. Women with type 1 diabetes have a higher risk of having a baby that is large for gestational age (LGA) or macrosomic [2]. The hypothesis that maternal hyperglycaemia results in fetal hyperinsulinaemia and consequently LGA or macrosomia is undisputed [3]. However, it is long recognised that maternal hyperglycaemia per se is not the only factor contributing to birthweight variation amongst offspring of women with type 1 diabetes. From clinical observation, even women with optimal glycaemic status can have offspring that are classified as LGA or macrosomic [4].

Placental weight correlates with neonatal weight at birth [5–7]. Recently, there has been renewed interest in placental growth factors (PlGFs) and their contribution to the development of preeclampsia and the consequent effect on fetal size. Fetal size is influenced by the fetoplacental circulatory system which begins forming in early gestation and develops throughout pregnancy. PlGFs contribute to the development of the fetoplacental circulatory system. PlGF is a pro-angiogenic factor secreted by the placenta, which reaches peak concentration at approximately 30 weeks of gestation and then declines [8]. PlGF exerts its pro-angiogenic effects largely through enhancing the effects of vascular endothelial growth factors (VEGFs) [9, 10]. Soluble fms-tyrosine kinase (sFlt-1) is a splice variant of Flt (a VEGF receptor) which acts

as an antiangiogenic factor by reducing free serum levels of both PlGF and VEGF [11].

Studies of placental factors have largely been in the context of preeclampsia, where low PlGF (<100 pg/ml) and/or a high sFlt-1/PlGF ratio (>85) are useful as predictors of impending preeclampsia [12]. Women who develop preeclampsia tend to have growth-restricted offspring. Studies in pregnant women without diabetes have also shown an association between low PlGF levels and infants that are small for gestational age (SGA), even in the absence of preeclampsia [13–16]. There are limited data on the association between PlGFs and birthweight *z* score in women with large babies, and very limited data on women with diabetes. Our aims were: first, to assess the association between PlGFs and birthweight *z* score in offspring of women with type 1 diabetes; and second, to determine if this association was altered by maternal glycaemic status.

Methods

Study design and population

This PlGF study was a secondary analysis of the Continuous Glucose Monitoring in Pregnant Women with Type 1 Diabetes (CONCEPTT) trial. The details of the CONCEPTT trial have been previously published [4]. In brief, CONCEPTT was a multicentre randomised controlled trial of real-time continuous glucose monitoring (CGM) in women with type 1 diabetes who were pregnant or planning pregnancy. Only women who were pregnant were included in this study. Pregnant women with type 1 diabetes aged between 18 and 40 years, with a duration of diabetes >1 year and under 14 weeks of gestation, were randomised to capillary glucose monitoring either with or without CGM. The primary outcome was the change in HbA_{1c} from randomisation to 34 weeks of gestation. In addition, other CGM measures of glycaemic control, such as percentage of time in range (%TIR (3.5–7.8 mmol/l [63–140 mg/dl]) and percentage of time above range (TAR) (% TAR > 7.8 mmol/l [140 mg/dl]), were obtained at baseline (approximately 12 weeks) and at 24 and 34 weeks of gestation in both groups. Women were eligible for this study if they had blood samples taken for PlGFs at approximately 12, 24 and 34 weeks of gestation.

Ethics

The CONCEPTT trial was approved by the Health Research Authority, East of England Research Ethics Committee (12/EE/0310) for all UK sites and at each individual centre for all other sites. All participants provided written, informed consent. The PlGF study was approved by the Mount Sinai Hospital Research Ethics Board.

Laboratory assays

For measurement of circulating PlGFs, blood was collected into BD Vacutainer Serum Separator Tubes (Becton Dickinson, USA), allowed to clot for 30 min at room temperature, centrifuged within 2 h of collection and transferred to cryovials for storage at −80°C prior to analysis. PlGF and sFlt-1 were analysed using commercially available ELISAs (R&D Systems, Minneapolis, MN, USA) according to the manufacturer's instructions. The interassay coefficients of variation were 7.2% and 5.6% for PlGF and sFlt-1, respectively. All HbA_{1c} measurements were performed at a central laboratory (Dynacare, Brampton, ON, Canada) using the turbidimetric inhibition immunoassay for haemolysed whole blood on the Cobas Integra 700 platform (Roche, Basel, Switzerland).

Definitions and outcome measures

Optimal glucose control was defined as $HbA_{1c} < 48$ mmol/mol (6.5%), and suboptimal glucose control as $HbA_{1c} \geq 48$ mmol/mol (6.5%), CGM percentage time above range (% TAR > 7.8 mmol/l [140 mg/dl]) was dichotomised as optimal (≤30% TAR) and suboptimal (>30% TAR). Gestational weight gain was calculated from randomisation to 34 weeks and used as a continuous variable. BMI was calculated from enrolment and used as a categorical variable divided into three categories: normal BMI <25 kg/m^2, overweight 25 to <30 kg/m^2, obese ≥30 kg/m^2.

We utilised high PlGF and a low sFlt-1/PlGF ratio as a surrogate for a 'healthy' placenta. Pathological injury to the placenta resulting in fetal growth restriction occurs via an ischaemia–reperfusion insult to the developing placenta cells. Persistent ischaemia–reperfusion damage to the placenta causes the histological features of maternal vascular malperfusion. This malperfusion ultimately results in an 'unhealthy placenta' and an excessive production and secretion of sFlt-1 with the suppression of secretion of PlGF. As demonstrated in the literature, abnormal circulation of sFlt-1 and PlGF correlates with the extent of placental malperfusion pathology [17, 18]. This was the basis of our rationale for using PlGF and sFlt-1/PlGF ratio to discriminate between healthy and unhealthy placentas. When we defined these angiogenic factors in a dichotomous fashion, a 'healthy' placenta had a PlGF of >100 pg/ml or sFlt-1/PlGF ratio of <85 and an 'unhealthy' placenta had a PlGF of <100 pg/ml or sFlt-1/PlGF ratio of >85 [12]. The cut-off for the sFlt-1/PlGF ratio was based on the study by Verlohren et al [19], whereby, with a cut-off of 85, the sFlt-1/PlGF ratio yields the highest sensitivity (89%) and specificity (97%) for early-onset

preeclampsia (pre 34 weeks of gestation). Whereas, for the late-onset preeclampsia group (>34 weeks of gestation), a cut-off of 85 results in a sensitivity of 74% and a specificity of 89%. The cut-off of 100 pg/ml for PlGF was based on a prospective cohort of 274 women with suspected preeclampsia, whereby a PlGF <100 pg/ml had high sensitivity to detect SGA births (sensitivity 93%, negative predictive value [NPV] 90%) when compared with fetal weight estimation by ultrasound (sensitivity 71%, NPV 79%) [20].

Birthweight *z* score was used as the primary outcome in this study. SGA was defined as birthweight <10th centile and LGA was defined as birthweight >90th centile using gestation-related optimal weight (GROW)-customised centiles [21].

The neonatal composite outcome was modified from CONCEPTT, and included birth injury, shoulder dystocia, neonatal hypoglycaemia, hyperbilirubinaemia, respiratory distress syndrome or neonatal care admission, but excluded pregnancy loss (miscarriage, stillbirth or neonatal death), as we were interested in the outcome of birthweight.

Statistical analysis

The main analyses were restricted to those with HbA_{1c}, PlGF and sFlt-1/PlGF ratio measured at 34 weeks and a livebirth infant. Scatterplots and Spearman correlations were used to assess univariate relationships of infant birthweight *z* score with PlGF and the sFlT-1/PlGF ratio after stratifying by maternal glycaemic control. Linear regression models were used to assess whether the relationship between birthweight *z* score and the growth factors differed according to maternal antenatal glycaemic control. One model was run for PlGF as the measure of placental health and another with the sFlT-1/PlGF ratio as the measure of placental health. In these models, birthweight *z* score was the outcome and the predictors were smoking, maternal BMI (modelled with a three-degree of freedom natural spline to account for nonlinearity), maternal weight gain, the logarithm of the growth factor, HbA_{1c} group (optimal $HbA_{1c} < 48$ mmol/mol [6.5%], suboptimal $HbA_{1c} \geq 48$ mmol/mol [6.5%] and an interaction between these terms. The interaction represents the difference between the optimal and suboptimal HbA_{1c} groups in the slope relating the birthweight *z* score to the PlGF. This analysis was repeated replacing HbA_{1c} by % TAR (>7.8 mmol/l [140 mg/dl]) as the measure of maternal antenatal glycaemic control, dichotomised as optimal (≤30% TAR) and suboptimal (>30% TAR). In exploratory analyses, the effects of maternal % TAR, HbA_{1c} and growth factors at baseline (12 weeks) and at 24 weeks were also examined. To quantify the relative importance of the predictors in the explanation of variance (R^2) in birthweight *z* scores, we used the Lindemann, Merenda and Gold method to partition the total unadjusted R^2 [22]. Interpretation of the PlGF–glycaemic interactions was facilitated by the use of box plots of birthweight *z* scores in four groups categorised as healthy/unhealthy placental function and optimal/suboptimal glucose control. All analyses used R version 3.6.1 [23].

Results

Baseline characteristics

The analytic cohort included 157 CONCEPTT participants with complete data for PlGF and sFlt-1 variables at 34 weeks of gestation and infant birthweight *z* score. We excluded 92 participants from the original cohort of 249 pregnant women in CONCEPTT [4]: one withdrew before baseline assessment could be obtained, 89 were missing bloods, and, of those 159 women with bloods, one was missing birthweight due to a withdrawal from the study and one had a stillborn infant, leaving 157 women in this cohort. Women who were included were similar to those excluded (electronic supplementary material [ESM] Table 1). The baseline characteristics of the PlGF study cohort are presented in Table 1. Baseline characteristics according to glycaemic measures can be found in ESM Table 2.

The results are presented in groups categorised as healthy or unhealthy PlGF levels (PlGF > or ≤100 pg/ml and sFlt-1/PlGF ratio > or ≤85). Of note, the duration of diabetes and HbA_{1c} at randomisation and at 34 weeks of gestation were similar across all groups. The percentage of women with microvascular complications also did not differ between the groups. Women with a low PlGF (and high ratio) had more gestational hypertension but less chronic hypertension, and rates of preeclampsia were similar across all groups. The percentage of infants categorised as SGA and LGA was similar across all groups when glycaemic control was not considered.

PlGF and infant birthweight *z* score by maternal glycaemic status

Overall, there was a small correlation between PlGF and infant birthweight *z* score (r = −0.08; 95% CI −0.08, 0.24, at 34 weeks of gestation). However, the slopes relating PlGF levels to birthweight *z* scores differed according to maternal glycaemia as assessed by HbA_{1c} at 34 weeks of gestation, after adjusting for maternal BMI, smoking and maternal weight gain ($p = 0.003$) (Fig. 1, ESM Table 3).

In the setting of optimal maternal glycaemia ($HbA_{1c} <$ 48 mmol/mol [6.5%] and % TAR ≤ 30%), birthweight *z* scores were reduced towards zero (normal weight) with increasing PlGF values (representing a healthy placenta). The mean birthweights and *z* scores were lower (birthweight = 3558 g; *z* score = 1.41) for those with a healthy placenta

Table 1 Baseline characteristics of the study population, dichotomised into high/low PlGF and sFLT/PlGF at 34 weeks of gestation

Characteristic	All women included	PlGF>100 pg/ml	PlGF≤100 pg/ml	sFLT-1/PlGF≤85	sFLT-1/PlGF>85
N	157	127	30	128	29
Age at entry (years)	31 (4.6)	31.2 (4.6)	33.1 (4.2)	31 (4.6)	33.5 (4)
BMI category, *n* (%)					
Normal (<25 kg/m^2)	82 (52.2)	64 (50.4)	18 (60)	66 (51.6)	16 (55.2)
Overweight (25 to <30 kg/m^2)	54 (34.4)	44 (34.6)	10 (33.3)	43 (33.6)	11 (37.9)
Obese (≥30 kg/m^2)	21 (13.4)	19 (15)	2 (6.7)	19 (14.8)	2 (6.9)
T1D duration (years), median (IQR)	15 (11–22)	15 (11–22)	16 (12–23)	15 (11–22)	16 (12–20)
Gestational age at birth (weeks)	37.2 (1.4)	37.3 (1.3)	36.5 (1.6)	37.3 (1.4)	36.6 (1.5)
HbA_{1c} at randomisation					
mmol/mol	51.5 (6.2)	51.6 (5.7)	51.5 (6.3)	51.4 (6.3)	51.8 (5.7)
%	6.9 (0.6)	6.9 (0.6)	6.9 (0.5)	6.9 (0.6)	6.9 (0.5)
HbA_{1c} at 34 weeks					
mmol/mol	47.0 (7.0)	48.1 (5.8)	46.7 (7.2)	46.6 (7.3)	48.5 (5.4)
%	6.5 (0.6)	6.4 (0.7)	6.6 (0.5)	6.4 (0.7)	6.6 (0.5)
Diabetes complications, *n* (%)					
Retinopathy	30 (19.1)	25 (19.7)	5 (16.7)	25 (19.5)	5 (17.2)
Nephropathy	2 (1.3)	2 (1.6)	0 (0)	2 (1.6)	0 (0)
Neuropathy	4 (2.5)	2 (1.6)	2 (6.7)	2 (1.6)	2 (6.9)
Chronic hypertension	5 (3.2)	5 (3.9)	0 (0)	5 (3.9)	0 (0)
Preeclampsia, *n* (%)	17 (10.8)	14 (11)	3 (10)	13 (10.2)	4 (13.8)
Gestational hypertension, *n* (%)	17 (10.8)	7 (5.5)	10 (33.3)	6 (4.7)	11 (37.9)
Birthweight (g)	3650 (648)	3688 (639)	3489 (673)	3677 (645)	3533 (660)
Birthweight *z* score	1.87 (1.52)	1.88 (1.41)	1.87 (1.55)	1.87 (1.54)	1.89 (1.43)
SGA[a], *n* (%)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
LGA[b], *n* (%)	102 (65.0)	82 (64.6)	20 (66.7)	83 (64.8)	19 (65.5)

Data are mean (SD) unless otherwise stated

[a] SGA is defined as birthweight *z* score <10th percentile as calculated using customised birthweight *z* score percentiles (GROW)

[b] LGA is defined as birthweight *z* score >90th percentile as calculated using customised birthweight *z* score percentiles (GROW)

T1D, type 1 diabetes

compared with those with an unhealthy placenta by PlGF (birthweight = 3619 g; *z* score = 1.93) (Fig. 2).

However, in women with suboptimal glycaemia (HbA_{1c} ≥ 48 mmol/mol [6.5%] and % TAR >30%), those with healthy placental function (higher PlGF) had heavier offspring (birthweight *z* scores) than those with lower PlGF (Figs 1, 2). In women with suboptimal glycaemia, the mean birthweights and *z* scores were higher (birthweight = 3853 g; *z* score = 2.45) for those with a healthy placenta than for those with an unhealthy placenta (birthweight = 3415 g; *z* score = 1.86) by PlGF.

When HbA_{1c} at 34 weeks is divided into three categories reflecting excellent, near-optimal and suboptimal glycaemia, as HbA_{1c} increases, the relationship between PlGF and birthweight *z* score increases (ESM Fig. 1). This difference in the slopes was not found for HbA_{1c} and PlGF levels at 12 and 24 weeks of gestation (data not shown). The contribution of these covariates to the unadjusted R^2 for birthweight *z* score was 18.7% overall, with BMI, smoking and gestational weight gain making up 8.9%, and the contribution of HbA_{1c}, log (PlGF) and the interaction of PlGF and HbA_{1c} group making up 9.8%. Similar relationships between PlGF levels and birthweight *z* score when stratified for glycaemic status were seen in preterm (<37 weeks) and term (≥37 weeks) infants at 34 weeks (ESM Fig. 2).

A similar difference was found in the slopes of the relationship between PlGF levels and birthweight *z* score when stratified by % TAR ≤ or >30%; $p = 0.049$ at 34 weeks (Fig. 1, ESM Table 4). This was not seen at 12 or 24 weeks of gestation (data not shown).

sFlt-1/PlGF ratio and infant birthweight *z* score by maternal glycaemic status

Similar to PlGF values, there was only a small correlation between sFlt-1/PlGF ratio and infant birthweight *z* score (r =

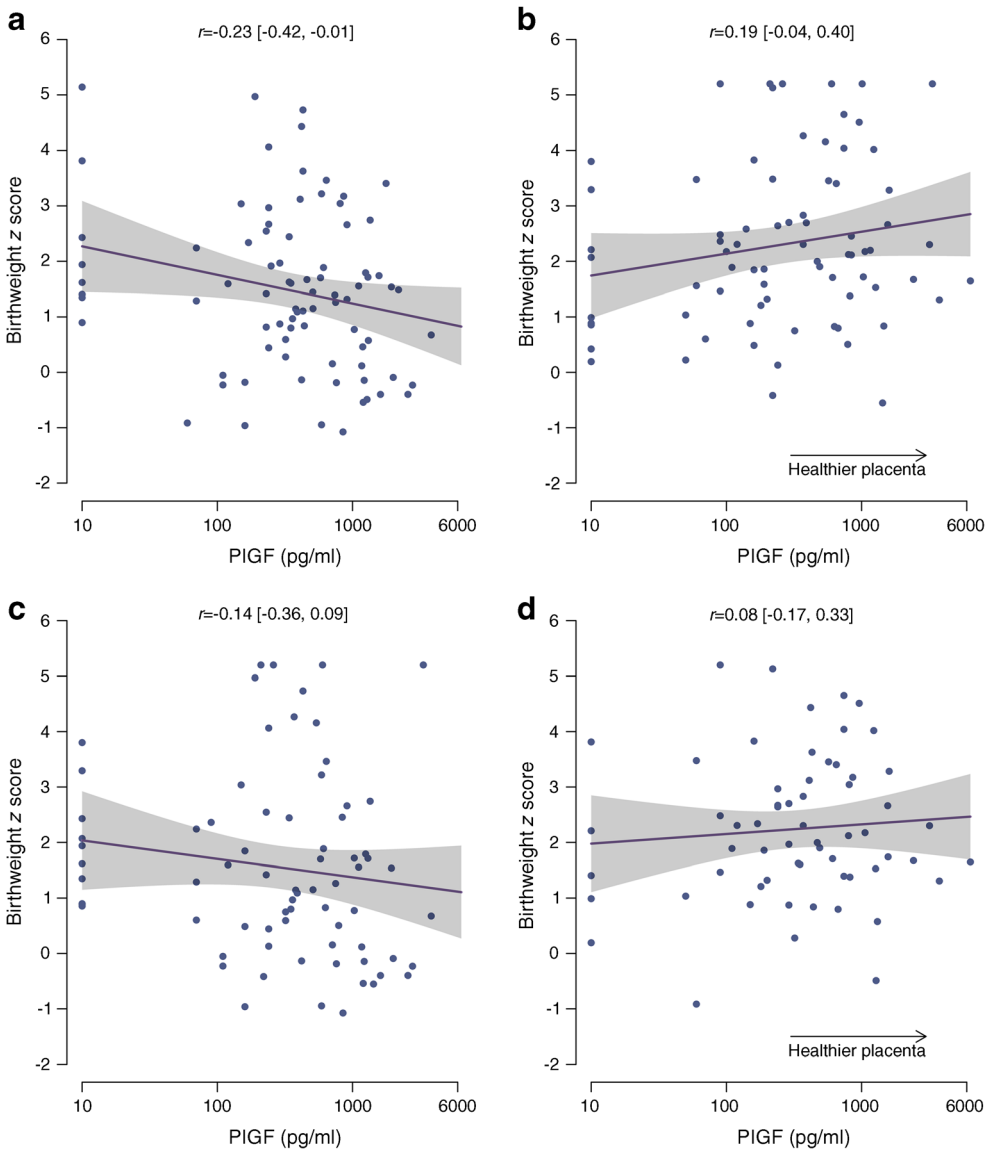

Fig. 1 Linear model of PlGF at week 34 and offspring birthweight *z* score by maternal glycaemic status with linear fit adjusted for maternal BMI, smoking and maternal weight gain. Maternal glycaemic status is defined as (**a**) optimal HbA_{1c} <48 mmol/mol or < 6.5% or (**b**) suboptimal HbA_{1c} ≥48 mmol/mol or ≥6.5%, and using % TAR 7.8 mmol/l (140 mg/dl) as (**c**) optimal ≤30% or (**d**) suboptimal >30%

0.18; 95% CI 0.01, 0.32, at 34 weeks of gestation). However, the slope of the relationship of the sFlt-1/PlGF ratio and mean birthweight *z* score differed significantly according to maternal glycaemic status by HbA_{1c} after adjusting for maternal BMI, smoking and gestational weight gain ($p = 0.0007$) (Fig. 3, ESM Fig. 3, ESM Table 5).

Similar to PlGF values alone, women with optimal glucose levels ($HbA_{1c} < 48$ mmol/mol [6.5%]) and healthy placental function (as measured by sFlt-1/PlGF ratio) had lighter babies (closer to normal weight) compared with those with unhealthy placentas (higher sFlt-1/PlGF ratio). In women with suboptimal glycaemic status ($HbA_{1c} \geq 48$ mmol/mol [6.5%]) those with healthier placentas as measured by a low sFlt-1/PlGF ratio had heavier offspring compared with those with an unhealthy placenta (see Fig. 3, ESM Fig. 3).

The slope of the relationship between the sFlt-1/PlGF ratio and birthweight *z* score also differed according to glycaemic status as measured by CGM measures (those with ≤30% time spent above the CGM hyperglycaemic target range of 7.8 mmol/l [140 mg/dl] vs those with >30% TAR) (Fig. 3, ESM Table 6) (difference in slopes $p = 0.003$). For those with near-optimal glycaemic status as evidenced by time above target <30%, infants were lighter when placentas were healthy. For those with suboptimal glycaemic control with >30% of time spent above range, infants were heavier when placentas were healthy (low sFlt-1/PlGF ratio) compared with those with an unhealthy placenta (high sFlt-1/PlGF ratio) (Fig. 3, ESM Fig. 3).

The concordance between the measures of placental function (PlGF and sFlt-1/PlGF ratio) was very good (κ [kappa statistic] = 0.85; 95% CI 0.75, 0.96).

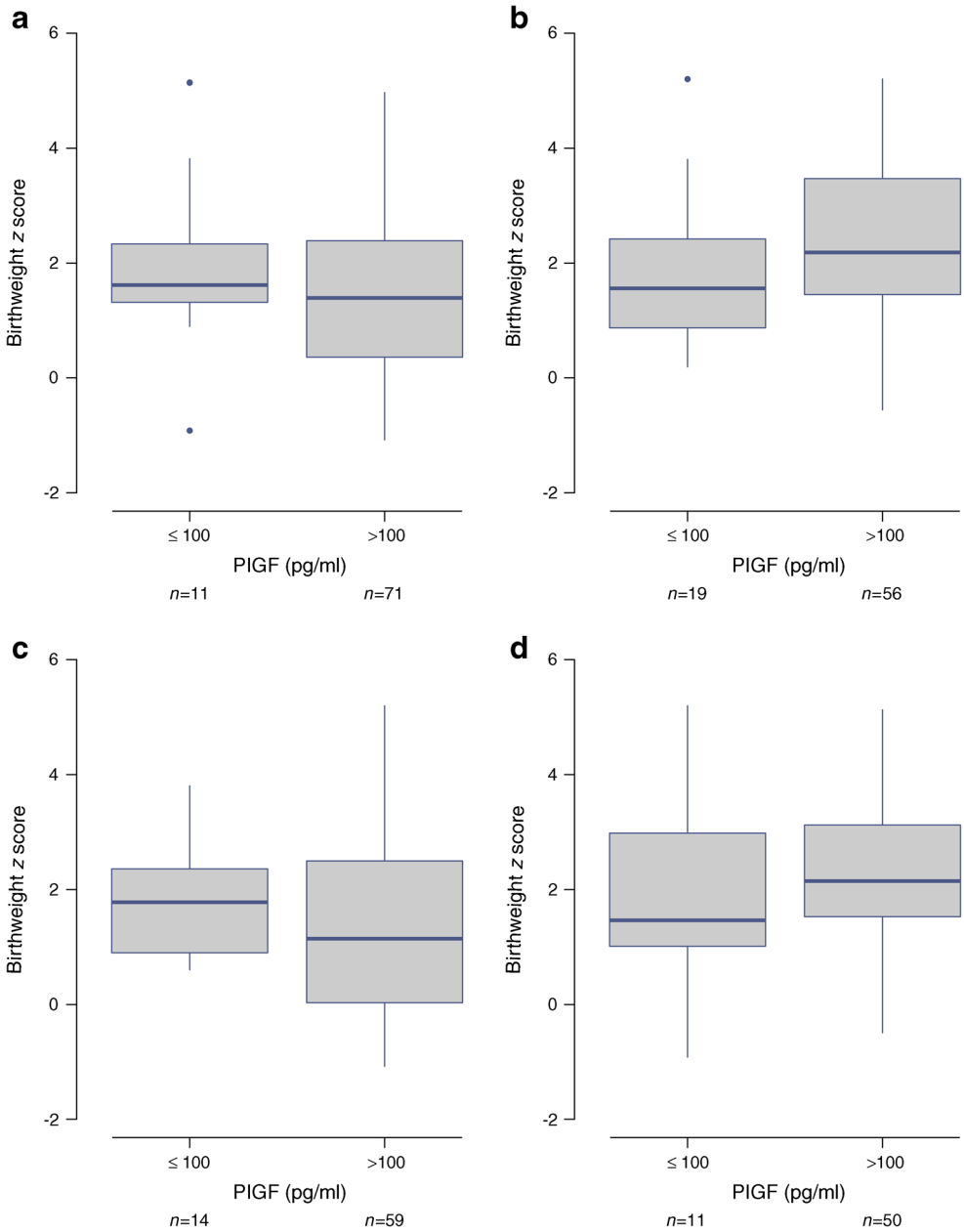

Fig. 2 Box plots depicting birthweight *z* scores according to low PlGF (≤100 pg/ml) vs high PlGF (>100 pg/ml) stratified by maternal glycaemic status defined as (**a**) optimal HbA_{1c} < 48 mmol/mol or <6.5% or (**b**) suboptimal HbA_{1c} ≥ 48 mmol/mol or ≥6.5% at 34 weeks of gestation, and using % TAR 7.8 mmol/l (140 mg/dl) as (**c**) optimal ≤30% or (**d**) suboptimal >30%

Neonatal outcomes

The neonatal composite outcome occurred in 7/19 (36.8%) of the high-HbA_{1c}, low-PlGF group (i.e., with suboptimal glycaemic status and unhealthy placental function) compared with 24/56 (42.9%) of the high-HbA_{1c}, high-PlGF group (i.e., with suboptimal glycaemic status and healthy placental function at 34 weeks of gestation) ($p = 0.85$).

Discussion

The offspring of women with type 1 diabetes and suboptimal glycaemic status had heavier birthweight in the setting of placental angiogenic factor levels considered 'healthy' (higher PlGF and lower sFlt-1/PlGF ratio). Conversely, infants of mothers with suboptimal glycaemic status, and placental angiogenic factors considered 'unhealthy' (lower PlGF and high sFlt-1/PlGF ratio), were lighter. The difference was apparent whether glycaemic status was assessed by conventional HbA_{1c} or by % TAR measures. In women with optimal glucose control the opposite was seen, with healthier placentas associated with lighter infants and unhealthy placental function associated with infants with larger birthweight.

What are the clinical implications of altered PlGFs?

PlGF as a predictor of macrosomia We found that the heaviest babies were those in mothers with poor glycaemic status and

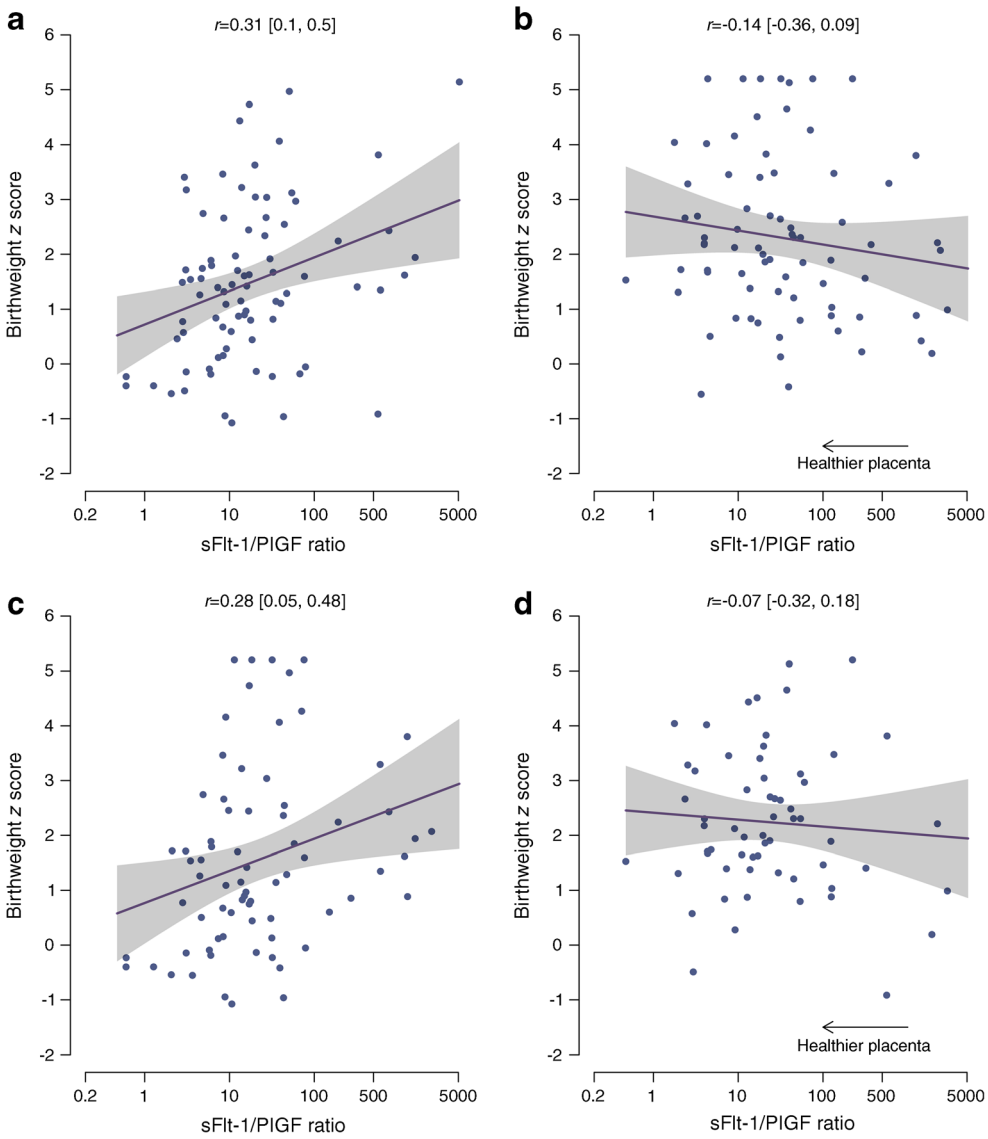

Fig. 3 Linear model of sFlt-1/PlGF ratio at week 34 and offspring birthweight *z* score by maternal glycaemic status with linear fit adjusted for maternal BMI, smoking and maternal weight gain. Maternal glycaemic status is defined as (**a**) optimal HbA_{1c} < 48 mmol/mol or <6.5% or (**b**) suboptimal HbA_{1c} ≥ 48 mmol/mol or ≥6.5%, and using % TAR 7.8 mmol/l (140 mg/dl) as (**c**) optimal ≤30% or (**d**) suboptimal >30%

healthy placentas. Transplacental glucose transfer is dependent upon not only the glucose concentration gradient from mother to fetus, but also the uteroplacental blood flow [24, 25]. PlGF, sFlt-1 and VEGF all regulate the development of the placental vascular system. We speculate that an elevation of PlGF (and/or a low sFlt-1/PlGF ratio) may increase placental vascularisation and increase glucose-transporting capacity and the transfer of other fuels, resulting in larger babies. On the other hand, reductions in PlGF (or elevations in sFlt-1/PlGF ratio) may result in placental dysfunction with reduced transfer of fuels and smaller babies. Several studies have shown a strong association between low PlGF levels and small babies in women without diabetes [14–16]. Few studies, however, have looked at these associations between PlGFs and birthweight in women with diabetes. Kuc et al found that normal levels of PlGF were associated with macrosomia, whereas lower levels were associated with infants of normal size, suggesting a role for PlGF in the size of the baby [26]. James-Todd et al found that third-trimester PlGF levels were significantly associated with birthweight and that high PlGF in the third trimester was associated with an increased risk of macrosomia [27]. They found no association between levels of PlGF and HbA_{1c} in women with type 1 (75%) and type 2 (25%) diabetes. It is unfortunate that levels of PlGF and sFit-1/PlGF ratio stratified by HbA_{1c} were not helpful earlier than 34 weeks. Further work in this area with larger sample sizes may allow better use of these angiogenic markers earlier in pregnancy.

To the best of our knowledge, no previous studies have looked at the association between placental function and infant birthweight stratified for maternal glycaemic status. We found that third-trimester PlGF levels were associated with a bigger baby in the context of suboptimal glucose status and a 'healthy' placenta. In women with suboptimal

glycaemic status and a healthy placenta (elevated PlGF or low sFlt-1/PlGF ratio), aiming for stricter glucose targets (HbA_{1c} < 42 mmol/mol [6%]) and less than 25% of time spent above the CGM target range (7.8 mmol/l [140 mg/dl]) may be beneficial in reducing accelerated fetal growth [28, 29]. In women with optimal glycaemic control, a healthy placenta was associated with a more normal birthweight *z* score, but an unhealthy placenta was associated with a larger baby. The cause for this is unknown.

PlGF and fetal growth restriction Not all women with suboptimal glycaemic status have a large baby. We have shown that in women with above-target HbA_{1c} level and low PlGF, infants were smaller. These babies, although not meeting clinical definitions of intrauterine growth restriction (IUGR < 2500 g) or SGA (<10th birthweight centile), are still potentially growth restricted in the setting of maternal hyperglycaemia. It has been shown that low third-trimester PlGF can distinguish placenta-mediated fetal growth restriction, detected post hoc by placental lesions indicative of under-perfusion [13]. This may be the case in our 'smaller' infants born to mothers with suboptimal glucose status and 'unhealthy' placentas as measured by angiogenic markers. Fetal growth restriction is associated with multiple neonatal complications including prolonged neonatal intensive care unit (NICU) admissions, hypoglycaemia, infection and respiratory distress [30].

In addition, low PlGF levels are associated with other adverse pregnancy outcomes in women without diabetes. In their meta-analysis, Sherrell et al found an association between low PlGF levels and Caesarean section for fetal compromise, NICU admission and stillbirth [13]. With only 19 infants in the high-HbA_{1c}, low-PlGF group, we did not have sufficient numbers to evaluate adverse neonatal effects. A larger cohort would be necessary to examine the effect of an 'unhealthy' placenta, particularly in normotensive women with type 1 diabetes. More frequent obstetric surveillance (for example, utilising Doppler artery studies) may be appropriate. More women in the 'unhealthy' placenta group had gestational hypertension, which may have been related to placental insufficiency and have played a role in the smaller babies. However, this effect was more prominent in those with suboptimal glucose status, suggesting that gestational hypertension may not be as detrimental on birthweight *z* score in women with optimal glycaemia.

Our study has several strengths. One is the well-characterised cohort of pregnant women with type 1 diabetes who were followed prospectively with detailed glycaemic measures using both HbA_{1c} and CGM. It was performed across 31 centres in six countries and data are therefore generalisable to many healthcare settings. We were able to evaluate PlGF and sFlt-1 in each trimester and to correlate with infant birthweight *z* score, stratified for maternal glycaemic status. There are also some limitations. We were unable to obtain blood samples from all CONCEPTT participants. With a larger sample size, we may have been able to explore whether the potential 'growth-restricted' infants had other neonatal complications and to assess the influence of different PlGF levels and sFlt-1/PlGF ratios on neonatal outcomes. More studies are needed to understand why women with optimal glycaemic control had poor placental health and bigger babies.

Conclusions In women with type 1 diabetes, infant birthweight is influenced both by maternal glycaemic status and by placental function, as measured by PlGF levels and sFlt-1/PlGF ratio. In the future, placental function assessment and additional obstetric surveillance may be appropriate for women with suboptimal glycaemic status and apparently normal fetal growth. Likewise, assessing longitudinal placental function measurements may also be applicable in women with optimal glycaemic status who are predicted to have babies with large birthweight. Measuring PlGF and/or sFlt-1/PlGF ratios in pregnant women with type 1 diabetes may help to predict compromised placental function and reduce perinatal complications. Although these results are intriguing, they are preliminary and future research will help to determine the usefulness of incorporating these measures of placental function into clinical practice outside of their use in predicting preeclampsia.

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Data availability All data generated or analysed during this study are included in this published article (and its supplementary information files).

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