Serum growth differentiation factor 15 is associated with glucose metabolism in the third trimester in Chinese pregnant women

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ARTICLE INFO

ABSTRACT

Objective: Growth differentiation factor 15 (GDF15) has been demonstrated to increase in diabetes as a protective factor. However, studies assessing relationships between GDF15 levels and gestational diabetes mellitus (GDM) are limited. In this study, we aimed to investigate whether GDF15 levels are related to GDM in Chinese subjects.

Methods: We included 200 GDM patients and 200 matched normal controls in the second trimester as well as 130 GDM patients and 130 matched normal controls in the third trimester. Serum GDF15 levels of all participants were determined using an enzyme-linked immunosorbent assay (ELISA). Then, according to GDF15 levels, we equally divided the participants in the second and third trimesters into four subgroups respectively. The relationships of serum GDF15 levels with glucolipid metabolism indicators were analyzed.

Results: In the third trimester, GDF15 levels were significantly higher in the GDM patients than in the normal controls ($P < 0.001$). Additionally, fasting blood glucose (FBG), 1-h postprandial glucose (1h-PG), 2-h postprandial glucose (2h-PG), hemoglobin A1C (HbA1c) and area under curve of glucose (AUCG) from the 75-g oral glucose tolerance test (OGTT) were positively associated with GDF15 levels ($P < 0.05$), even after adjusting for age, pregestational BMI, changes of BMI until the third trimester, gestational age, twin and family history of diabetes. Moreover, GDF15 levels were higher in the third trimester than in the second trimester ($P < 0.001$). No significant relationships were found between GDF15 levels and glucolipid metabolism in the second trimester ($P > 0.05$).

Conclusions: Serum GDF15 levels were positively correlated with glucose metabolism in the third trimester in Chinese pregnant women.

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1.   Introduction

Gestational diabetes mellitus (GDM), a disease defined as abnormal glucose tolerance with onset or first recognition during gestation [1], occurs in approximately 14.8% of all pregnancies in mainland China according to a recent meta-analysis [2]. GDM has been proved to be associated with a variety of complications, including gestational hypertension, cardiovascular disease and cesarean delivery in mothers as well as macrosomia, respiratory distress syndrome and hypoglycemia in neonates [3,4]. Moreover, GDM patients and their offspring will have a higher risk of type 2 diabetes (T2DM) and obesity in the long term [5]. To date, the pathogenesis of GDM is still unclear, but there is evidence that GDM is analogical to T2DM because of the similarity of the underlying pathophysiology of inadequate beta cell compensation to the increased insulin requirements and a majority of shared susceptibility genes [1,6].

Growth differentiation factor 15 (GDF15), also known as macrophage inhibitory cytokine-10 (MIC-1), is a distant member of the transforming growth factor β superfamily [7]. GDF15 is expressed in large amounts by placental trophoblasts and can be robustly induced by both acute and chronic inflammatory states, including tissue injury, cancer, cardiovascular disease, nephrosis and diabetes [8–11]. In recent years, significant associations between GDF15 and glycometabolic disorders have been found in human and animal studies. Temporary high glucose and insulin states could induce the transient increasement of GDF 15 levels [12,13], and GDF15 overexpressing mice had obviously reduced body weight and glucose intolerance as well as improved insulin sensitivity while an antibody to GDF15 could efficiently reverse these effects [14]. In addition, GDF15 was also demonstrated to play a significant role in regulating lipid levels by these findings that GDF15 levels were negative associated with total cholesterol and transgenic GDF15 overexpressing mice had improved lipid metabolism [9,15]. These evidences indicated GDF15 might be a protective factor in glucolipid metabolism.

The immune-tolerant state of mothers is important for a healthy pregnancy, so the immunosuppressive function of GDF15 might play a significantly protective role throughout gestation [7,16]. Study has shown that GDF15 levels were high in the serum of pregnant women, and its level increased substantially with the progression of gestation and peaked in the third trimester [8]. In view of the important role of GDF15 in gestation and glucolipid metabolism, there probably be some relationships between serum GDF15 levels and gestational diseases especially GDM. However, until now, only one study with a small sample size concluded that GDF15 levels in the second trimester were not significantly different between the patients with GDM and the normal controls [17]. Therefore, we aimed to investigate whether serum GDF15 levels in the second and third trimesters were associated with glucose and lipid metabolism parameters.

2.   Materials and methods

2.1.   Study subjects

Paired by maternal age (±3 years), pregestational BMI (±3 kg/m^2), and gestational week (±3 weeks), a total of 330 GDM patients and 330 matched normal controls were recruited from University of Hong Kong Shenzhen Hospital in this study, including 200 GDM patients and 200 matched normal controls in the second trimester as well as 130 GDM patients and 130 matched normal controls in the third trimester. Women with autoimmune disease, thyroid disease, heart trouble, liver or kidney disease, tumors, hematopathy, and other known diseases affecting glucolipid metabolism or serum levels of GDF15 were excluded from this study. A 75-g oral glucose tolerance test (OGTT) was performed for each subject in the second trimester (24–28 weeks) and GDM was diagnosed by the World Health Organization guidelines published in 2013 (fasting blood glucose (FBG) $\geq$ 5.1 mmol/L or 1-h postprandial glucose (1 h-PG) $\geq$ 10.0 mmol/L or 2-h postprandial glucose (2 h-PG) $\geq$ 8.5 mmol/L). Written informed consent has been obtained from each participant before undertaking this study. This study was approved by The Medical Ethics Committee of University of Hong Kong Shenzhen Hospital and conformed to the provision of the Declaration of Helsinki.

2.2.   Clinical measurements

Physical parameters were collected from each subject. Pregestational BMI was calculated as the pregestational weight in kilograms divided by the square of the height in meters. The blood samples of all participants were collected when the participants were in a resting state through the antecubital vein after an 8-h overnight fast. Glucose levels were measured using hexokinase method on a Roche Cobas 701 analyzer (Roche, Ltd., Basel, Switzerland). HbA1c was determined using a high-performance liquid chromatography method on an Arkray HA-8160 analyzer (Arkray, Ltd., Kyoto, Japan). Insulin was detected using a MILLIPLEX® MAP kit (Merck Corp., New Jersey, USA) on a MAGPIX analyzer (Merck Corp.). Lipid indexes were measured on a Siemens ADVIA2400 fully automated chemistry analyzer (Siemens AG, Munich, Germany). The area under curve of glucose from the 75-g OGTT (AUCG) was calculated as 1/2*[FBG (mmol/L) + 1 h-PG (mmol/L)] × 1 h + 1/2*[1 h-PG (mmol/L) + 2 h-PG (mmol/L)] × 1 h. The homeostatic model assessment index of insulin resistance (HOMA-IR) was calculated as FBG (mmol/L) × fasting insulin (mU/L)/22.5. The homeostatic model assessment index of β-cell secretion (HOMA-β) was calculated as 20 × fasting insulin (mU/L) / [FBG (mmol/L)-3.5].

2.3.   Serum GDF15 measurement

Serum GDF15 levels were measured using an ELISA kit (R&D Systems, Inc. California, USA) according to the manufac-

turer's instructions. This assay had high sensitivity to GDF15 with minimum detectable dose of human GDF-15 ranged from 0.0 to 4.4 pg/mL. Excellent specificity of this assay to GDF15 was also proved by nonsignificant cross-reactivity or interference with other GDF family members. The intra-assay variation ≤2.8% and inter-assay variation ≤6.0%.

2.4. Statistical analysis

All analyses were performed with SAS version 8.0 (SAS Institute, Cary, NC). GraphPad Prism software (version 7.0) was used to plot the histograms and scatterplots. Normally distributed data was expressed as the mean ± SD or as medians with the interquartile ranges. Student's paired t test or sign-rank test was used for comparison between two groups. The chi-square test was used for categorical variables. Subgroups were analyzed by variance analysis or Kruskal-Wallis test. Associations of serum GDF15 levels with glucose and lipid metabolism parameters were estimated by Spearman or Pearson correlation coefficients according to the distribution of relevant variables. Multiple regression analysis was used to examine the associations of log10-transformed serum GDF15 levels and other parameters with adjustments for maternal age, pregestational BMI, changes of BMI until the third trimester, gestational age, twin and family history of diabetes. A two-tailed $P < 0.05$ was considered statistically significant.

3. Results

3.1. Characteristics of subjects

Table 1 shows the clinical characteristics of all participants. In the third trimester, GDM patients were more likely to have higher changes of BMI and higher levels of FBG, 1 h-PG, 2 h-PG, AUCG, HbA1c, and gestational age (all $P < 0.05$), but lower total cholesterol, triglycerides and delivery gestational age than normal controls (all $P < 0.005$). In the second trimester, GDM patients also had higher levels of FBG, 1 h-PG, 2 h-PG, AUCG, fasting insulin, HOMA-IR, HbA1c, and HDL-C (all $P < 0.05$), but lower delivery gestational age, neonatal height and neonatal weight than normal pregnant women (all $P < 0.005$). There were no significant differences in maternal age and pregestational BMI between subjects in the second and third trimesters both for GDM and NGT (normal glucose tolerance) groups (all $P > 0.05$).
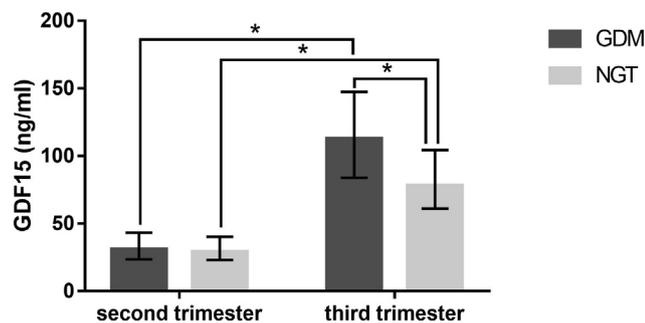
3.2. Serum GDF15 levels in GDM and NGT groups

Serum GDF15 levels in GDM patients were significantly higher than in the normal controls in the third trimester (112.78 ng/ml (83.87, 147.40) vs 78.13 ng/ml (61.04, 104.47)) ($P < 0.001$), but not in the second trimester ($P > 0.05$) (Fig. 1). We also found that serum GDF15 levels were higher in the third trimester than in the second trimester for both GDM patients and normal controls (cases: $P < 0.001$, controls: $P < 0.001$).

Table 1 – Characteristics of the GDM and NGT groups in the second and third trimesters.

Characteristics	Subjects in the second trimester			Subjects in the second trimester		
	GDM (n = 130)	NGT (n = 130)	P	GDM (n = 200)	NGT (n = 200)	P
Age (years)	31 (29,34)	31 (29,33)	0.897	32 (29,35)	32 (29,35)	0.906
Pregestational BMI (kg/m^2)	21.19 ± 2.29	21.06 ± 2.15	0.695	21.32 ± 2.59	21.26 ± 2.52	0.343
Changes of BMI (kg/m^2)	5.15 ± 1.66	5.78 ± 1.35	0.001	2.69 ± 1.23	2.70 ± 1.23	0.792
Family history of diabetes, n (%)	37 (18.5)	23 (11.5)	0.039	48 (24)	5 (2.5)	<0.001
Gestational age (weeks)	37 (36,37)	36 (36,37)	0.024	26 (25,27)	25 (25,26)	0.498
FBG (mmol/L)	4.70 ± 0.54	4.37 ± 0.28	<0.001	4.64 ± 0.53	4.36 ± 0.28	<0.001
1 h-PG (mmol/L)	9.71 ± 1.61	7.16 ± 1.43	<0.001	9.88 ± 1.32	7.45 ± 1.24	<0.001
2 h-PG (mmol/L)	8.55 ± 1.42	6.34 ± 1.01	<0.001	9.07 ± 1.24	6.66 ± 0.98	<0.001
AUCG (mmol/L h)	16.33 ± 2.07	12.52 ± 1.77	<0.001	16.74 ± 1.72	12.97 ± 1.56	<0.001
Fasting insulin (mU/L)				6.79 (4.75,10.78)	5.37 (3.73,7.77)	0.001
HOMA-IR				1.38 (0.95,2.21)	1.05 (0.71,1.53)	<0.001
HOMA-β				134.07 (88.68,206.35)	123.20 (81.80,215.58)	0.522
HbA1c (%)	5.40 (5.20,5.60)	5.20 (5.00,5.40)	<0.001	5.20 (5.00,5.40)	5.10 (5.00,5.30)	0.003
Total cholesterol (mmol/L)	6.18 ± 1.14	6.98 ± 1.20	<0.001	5.93 ± 1.03	6.07 ± 1.00	0.176
Triglycerides (mmol/L)	2.83 (2.10,3.90)	3.42 (2.73,4.11)	0.001	2.24 (1.85,2.81)	2.25 (1.80,2.85)	0.082
LDL-C (mmol/L)	3.45 ± 0.88	3.62 ± 0.85	0.117	3.45 ± 0.91	3.30 ± 0.77	0.085
HDL-C (mmol/L)	1.85 ± 0.36	1.93 ± 0.39	0.098	2.01 ± 0.43	1.87 ± 0.35	<0.001
Delivery gestational age (weeks)	39 (38,39)	39 (38,40)	0.030	39 (38,39)	39 (38,40)	0.002
Neonatal weight (kg)	3.16 ± 0.43	3.38 ± 0.36	0.668	3.16 ± 0.43	3.29 ± 0.44	0.003
Neonatal length (cm)	50 (49,50)	50 (50,50)	0.812	50 (49,50)	50 (50,50)	0.004

Data are means ± SD or median (interquartile range).
Abbreviations: NGT, normal glucose tolerance; BMI, body mass index; FBG, fasting blood glucose; 1h-PG, 1-h postprandial glucose; 2h-PG, 2-h postprandial glucose; AUCG, area under curve of glucose from the 75-g OGTT; HOMA-IR, homeostasis model assessment of insulin resistance; HOMA-β, homeostasis model assessment index of β-cell secretion; HbA1c, hemoglobin A1C; LDL-c, low-density lipoprotein cholesterol; HDL-c, high-density lipoprotein cholesterol.

Fig. 1 – Serum GDF15 levels in subjects in the second trimester with GDM (n = 200) and NGT (n = 200) as well as in the third trimester with GDM (n = 130) and NGT (n = 130). Median (interquartile range) serum levels of GDF15 in subjects with GDM and NGT are displayed. *, P < 0.001.

3.3. Association of serum GDF15 levels with clinical metabolism parameters

According to GDF15 levels, we divided the subjects in the second and third trimesters into four subgroups respectively, with an equal number of women in each subgroup. In the third trimester, 1 h-PG, 2 h-PG and AUCG increased with increasing quartiles of serum GDF15, and triglycerides were different among the four subgroups but without a clear trend (Table 2). Then, we conducted correlation analysis between log10-transformed GDF15 levels and clinical indexes. Results showed that FBG, 1 h-PG, 2 h-PG, AUCG and HbA1c were positively associated with GDF15 levels, even after adjusting for age, pregestational BMI, changes of BMI until the third trimester, gestational age, twin and family history of diabetes (all

$P < 0.05$) (Fig. 2). However, no significant differences were detected among the four subgroups in the second trimester ($P > 0.05$) (Table 3).

4. Discussion

Although there is emerging evidence from a great number of studies which have proved the important role of GDF15 in glucose and lipid metabolism [12,15], the clinical relationship between GDF15 and GDM has been seldom researched. In this study, we explored the associations between GDM and serum GDF15 levels and found that concentrations of GDF15 were positively correlated with glucose metabolism in the third trimester.

Serum GDF15 levels have been reported to be high in pregnant women and increase with the progressing of gestation [8,18]. The results of our research showed a similar tendency that GDF15 levels were higher in the third trimester than in the second trimester in both GDM patients and normal controls, whereas GDF15 levels of normal participants in our study were higher than those that have been reported [8,18]. The diverse detection methods and the very small sample sizes of previous studies might account for the differences. Although our finding was also in accordance with a previous study that GDF15 levels in normal pregnant women were not different from the levels of women with GDM in the second trimester [17], a remarkable difference was observed in the third trimester in our research, which indicated that GDF15 probably play an important role in the third trimester instead of in the second trimester. Some researchers have found that a short-term nutritional imbalance did not trigger long-term GDF15 production in both humans and mice, whereas

Table 2 – Relationships of clinical characteristics across GDF15 quartiles in the third trimester.

Charicteristics	Quartiles of serum GDF15 concentration				P
	Quartile 1 (≤68.95 ng/mL)	Quartile 2 (68.95–94.90 ng/mL)	Quartile 3 (94.90–126.00 ng/mL)	Quartile 4 (≥126.00 ng/mL)	
n	65	65	65	65	
Age (years)	31 (29,33)	31 (29,33)	31 (29,34)	31 (28,33)	0.660
Pregestational BMI (kg/m^2)	21.45 ± 2.16	21.05 ± 2.21	21.22 ± 2.30	20.78 ± 2.21	0.222
Changes of BMI (kg/m^2)	5.66 ± 1.43	5.68 ± 1.54	5.12 ± 1.60	5.38 ± 1.52	0.131
Gestational age (week)	37 (36,37)	37 (36,37)	36 (36,37)	37 (36,37)	0.353
FBG (mmol/L)	4.42 ± 0.40	4.53 ± 0.38	4.54 ± 0.46	4.64 ± 0.56	0.062
1 h-PG (mmol/L)	7.67 ± 1.57	8.30 ± 2.05	8.60 ± 1.90	9.18 ± 2.11	<0.001
2 h-PG (mmol/L)	6.80 ± 1.18	7.25 ± 1.55	7.53 ± 1.68	8.22 ± 1.85	<0.001
AUCG (mmol/L h)	13.28 ± 2.00	14.18 ± 2.67	14.63 ± 2.61	15.61 ± 2.99	<0.001
HbA1c (%)	5.20 (5.10,5.40)	5.30 (5.10,5.50)	5.20 (5.10,5.50)	5.40 (5.20,5.60)	0.053
Total cholesterol (mmol/L)	6.75 ± 1.12	6.66 ± 1.39	6.45 ± 1.17	6.44 ± 1.25	0.398
Triglycerides (mmol/L)	3.43 ± 1.62	3.51 ± 1.20	3.05 ± 1.29	3.60 ± 1.84	0.034
LDL-C (mmol/L)	3.50 ± 0.77	3.57 ± 0.96	3.54 ± 0.85	3.53 ± 0.88	0.947
HDL-C (mmol/L)	1.96 ± 0.37	1.87 ± 0.44	1.91 ± 0.39	1.83 ± 0.29	0.181
Delivery gestational age (weeks)	39 (39,40)	39 (38,40)	39 (39,40)	39 (38,39)	0.056
Neonatal weight (kg)	3.35 ± 0.37	3.40 ± 0.40	3.28 ± 0.38	3.39 ± 0.37	0.531
Neonatal length (cm)	50 (50,50)	50 (49,50)	50 (50,50)	50 (49,50)	0.243

Data are means ± SD or median (interquartile range).
Abbreviations: BMI, body mass index; FBG, fasting blood glucose; 1h-PG, 1-h postprandial glucose; 2h-PG, 2-h postprandial glucose; AUCG, area under curve of glucose from the 75-g OGTT; HbA1c, hemoglobin A1C; LDL-c, low-density lipoprotein cholesterol; HDL-c, high-density lipoprotein cholesterol.
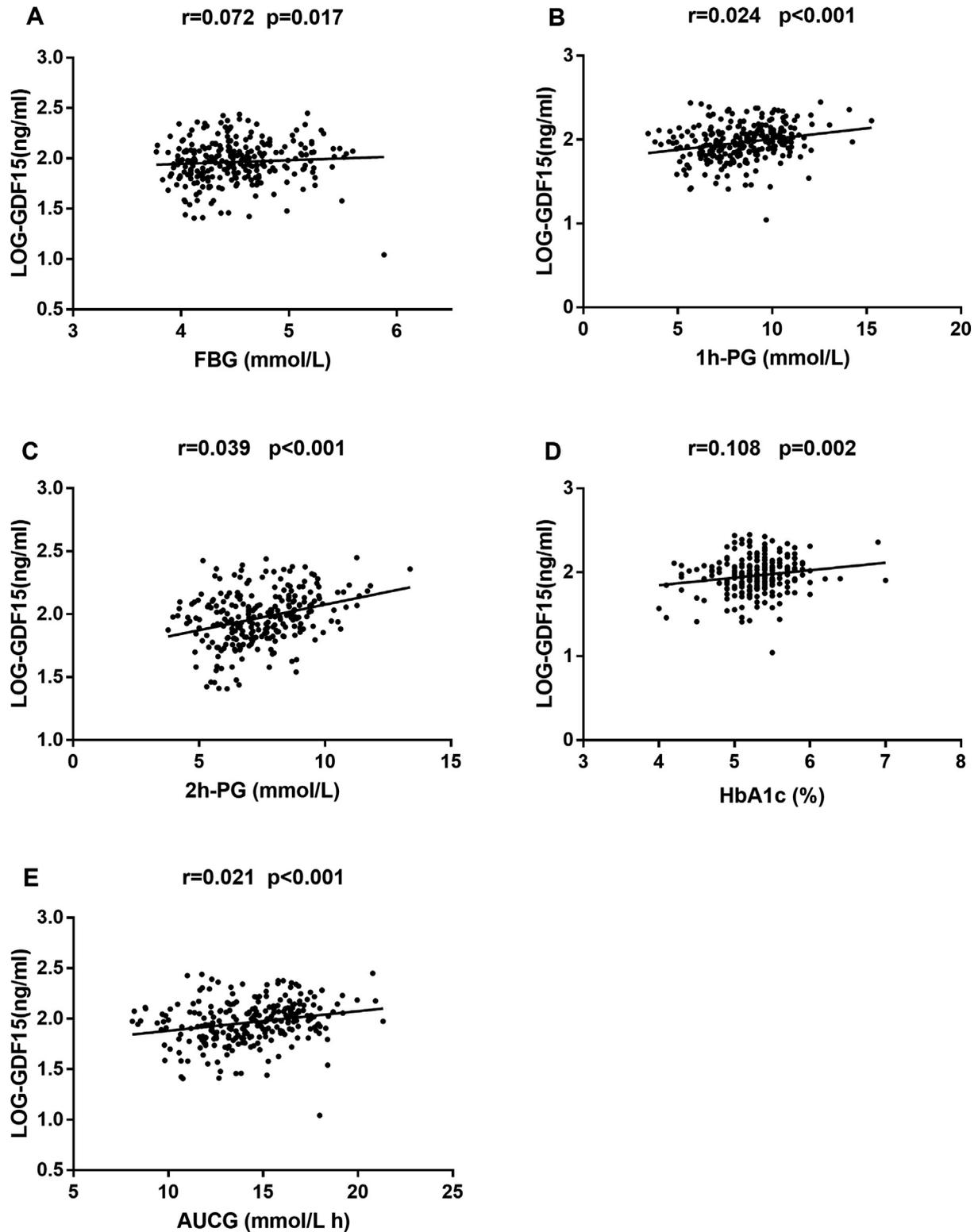
Fig. 2 – Correlations of serum levels of GDF15 (log-transformed) with 1h-PG, 2h-PG, HbA1c and AUCG in 260 pregnant women in the third trimester. r and p, adjusted for maternal age, pregestational BMI, changes of BMI until the third trimester, gestational age, twin and family history of diabetes.

prolonged nutritional stressors distinctly gave rise to an stably increase in GDF15 levels [19]. Therefore, we assumed it similarly took time for GDF15 to show a large difference and come into play in pregnancies with abnormal glucose levels.

The expression and secretion of GDF15 can be temporarily induced by high glucose levels [12,20]. Human studies also found GDF15 levels were positively associated with blood glucose [21,22]. The same tendency was shown in our study that

Table 3 – Relationships of clinical characteristics across GDF15 quartiles in the second trimester.

Charicteristics	Quartiles of serum GDF15 concentration				P
	Quartile 1 (≤23.20 ng/mL)	Quartile 2 (23.20–30.41 ng/mL)	Quartile 3 (30.41–42.00 ng/mL)	Quartile 4 (≥42.00 ng/mL)	
n	100	100	100	100	
Age (years)	31 (29,34)	32 (29,35)	32 (29.5,35)	32 (30,35)	0.735
Pregestational BMI (kg/m^2)	21.18 ± 2.09	21.38 ± 2.63	21.44 ± 2.65	21.16 ± 2.81	0.659
Changes of BMI (kg/m^2)	2.51 ± 1.35	2.62 ± 1.05	2.66 ± 1.21	2.97 ± 1.26	0.056
Gestational age (week)	25 (24,26)	25 (24,26)	25 (24,26)	25 (24,26)	0.505
FBG (mmol/L)	4.48 ± 0.56	4.50 ± 0.37	4.52 ± 0.40	4.49 ± 0.45	0.319
1 h-PG (mmol/L)	8.59 ± 1.70	8.69 ± 1.79	8.68 ± 1.84	8.70 ± 1.75	0.963
2 h-PG (mmol/L)	7.85 ± 1.73	7.72 ± 1.52	7.90 ± 1.68	7.98 ± 1.64	0.696
AUCG (mmol/L h)	14.75 ± 2.51	14.81 ± 2.48	14.91 ± 2.59	14.93 ± 2.44	0.951
Fasting insulin (mU/L)	7.35 (5.25,12.16)	6.26 (4.43,8.82)	5.67 (3.67,8.43)	5.06 (3.73, 7.16)	0.914
HOMA-IR	1.17 (0.84,1.87)	1.20 (0.83,1.92)	1.16 (0.71,1.81)	1.22 (0.83,1.77)	0.975
HOMA-β	150.16 (80.41,238.25)	125.58 (80.33,200.72)	112.97 (72.26,207.88)	129.36 (86.22,213.29)	0.554
HbA1c (%)	5.10 (5.0,5.3)	5.20 (5.00,5.30)	5.20 (5.00,5.30)	5.10 (5.00,5.30)	0.888
Total cholesterol (mmol/L)	6.04 ± 1.13	6.02 ± 1.00	5.93 ± 0.87	6.03 ± 1.07	0.876
Triglycerides (mmol/L)	2.30 (1.74,2.72)	2.13 (1.72,2.74)	2.26 (1.70,2.92)	2.30 (1.94,2.85)	0.326
LDL-C (mmol/L)	3.37 ± 0.85	3.34 ± 0.76	3.34 ± 0.75	3.37 ± 0.84	0.978
HDL-C (mmol/L)	1.93 ± 0.38	1.99 ± 0.41	1.91 ± 0.39	1.93 ± 0.43	0.313
Delivery gestational age (weeks)	39 (38,40)	39 (38,39)	39 (38,39)	39 (37,39)	0.347
Neonatal weight (kg)	3.27 ± 3.80	3.24 ± 4.37	3.28 ± 4.15	3.16 ± 5.07	0.251
Neonatal length (cm)	50 (50,50)	50 (50,50)	50 (50,50)	50 (49,50)	0.343

Data are means ± SD or median (interquartile range).

Abbreviations: BMI, body mass index; FBG, fasting blood glucose; 1 h-PG, 1-h postprandial glucose; 2 h-PG, 2-h postprandial glucose; AUCG, area under curve of glucose from the 75-g OGTT; HOMA-IR, homeostasis model assessment of insulin resistance; HOMA-β, homeostasis model assessment index of β-cell secretion; HbA1c, hemoglobin A1C; LDL-C, low-density lipoprotein cholesterol; HDL-C, high-density lipoprotein cholesterol.

serum GDF15 levels in the third trimester were positively associated with blood glucose and HbA1c even after adjustment for confounders. However, we didn't find the same relationships in the second trimester, which might be due to the reason we just discussed that it needed time for the obvious increasement of GDF15 levels. Studies have reported that inflammation could improve the expression of GDF 15, and the increasement of GDF15 was necessary for anti-inflammation cytokines to balance blood glucose through signal transducer and activator of transcription 6 (STAT6) signaling pathway [23]. Moreover, reduced glucose tolerance was shown in mice with GDF15-deficient macrophages owing to the changes in immuno-microenvironment of white adipose tissue [24]. Additionally, serum GDF15 levels were positively associated with the metformin dosage in people with abnormal glucose tolerance, suggesting the signaling pathways which regulated expression and secretion of GDF15 could be adjusted by metformin to control glucose homeostasis [25,26]. Based on the evidence above, we speculated GDF15 was a protective factor for glucose metabolism. However, the exact mechanism of GDF15 in patients with glucose metabolism disorders is still unclear and need further exploration.

We did not find any relationships between GDF15 levels and insulin secretion or sensitivity in the second trimester, which contradicted to some reports. Other researchers found that insulin sensitivity was improved in GDF15 transgenic mice as a result of reduced NLRP3 inflammasome activity in adipose tissue [27], and positive associations between insulin resistance and GDF15 levels were found in obese people [21]. We assumed our different results might be due to the differ-

ent physiological status between normal diabetes and GDM and the assumption we discussed previously that it took time for GDF15 to make a big difference and come into play. We did not have data regarding insulin secretion or sensitivity in the third trimester because some of these GDM patients had already been treated with insulin, but we suspected the relationship probably existed in the third trimester as there were close relationships between serum GDF15 and glucose metabolism.

Transgenic GDF15 overexpressing mice on both normal and high fat diet exhibited improved lipid metabolism through the increasement of thermogenesis, lipolysis and oxidation [15]. In addition, the expression of hepatic GDF15 could be induced by X-box-binding protein 1 (XBP1) signaling pathway in fasting state, which further promote fatty acid β-oxidation to decrease lipid accumulation [28]. Moreover, human studies have revealed GDF15 levels were negatively associated with total cholesterol and HDL cholesterol [9]. All of these data suggested that GDF15 acts as a protective factor in lipid disorders. However, in our study, no relationships between GDF15 and lipid metabolism indexes were found in either the second or third trimesters. One of the reasons we considered was that almost all GDM patients diagnosed in the second trimester had been treated with diet, exercise or insulin so that we could not see obvious dyslipidemia in the third trimester not to mention the relationships of lipid indexes and GDF15 levels.

There were still limitations in our study. First, because it was a case-control study, we could not examine the exact changes in serum GDF15 levels throughout the entire preg-

nancy or determine whether GDF15 could be a useful predictive factor for the diagnosis of GDM. However, our samples were from the second and third trimesters, which might offset some of the effects. Next, our sample size in the third trimester was relatively small, but we still found significant differences between cases and controls in our results, which indicated GDF15 does play a crucial role during the third trimester. Finally, we didn't have data relating blood glucose in the third trimester except the HbA1c, the correlation analysis was conducted between GDF15 and glucose levels which detected during 24 and 28 gestational weeks. However, in consideration of the close relationships of GDF15 and HbA1c, we have reasons to believe the important role of GDF15 in glucose metabolism in the third trimester. Further prospective and large sample size studies are still needed to explore the exact impacts of GDF15 on GDM patients throughout pregnancy.

In conclusion, our study using serums from two different periods provided clinical evidence demonstrating that GDF15 levels were positively correlated with glucose metabolism in the third trimester and might be a potential protective factor for gestational diabetes.

Acknowledgments

Author contributions

CH designed the study. MT performed the experiments, statistical analysis and wrote the manuscript. MT and ML collected and checked the data; WL, SW and RZ helped to perform the experiment. XY and XZ reviewed the manuscript. CH reviewed and revised the manuscript. All authors have read and approved the final manuscript.

Declaration of Competing Interest

The authors have nothing to disclose.

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