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Interleukin 6 (IL-6) and Tumor Necrosis Factor α (TNF-α) Single Nucleotide Polymorphisms (SNPs), Inflammation and Metabolism in Gestational Diabetes Mellitus in Inner Mongolia

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Statistical Analysis C
Data Interpretation D
Manuscript Preparation E
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Background: Gestational diabetes mellitus (GDM) is common all over the world. GDM women are with inflammatory and metabolisms abnormalities. However, few studies have focused on the association of IL-65–72C/G and TNF-α -857C/T single nucleotide polymorphisms (SNPs), inflammatory biomarkers, and metabolic indexes in women with GDM, especially in the Inner Mongolia population. The aim of this study was to investigate the associations of IL-65–72C/G and TNF-α -857C/T SNPs, and inflammation and metabolic biomarkers in women with GDM pregnancies.

Material/Methods: Blood samples and placentas from 140 women with GDM and 140 women with healthy pregnancies were collected. Matrix-assisted laser desorption ionization time of flight mass spectrometry (MALDI-TOF-MS) and MassARRAY-IPLEX were performed to analyze IL-65–72C/G and TNF-α -857C/T SNPs. Enzyme linked immunosorbent assay (ELISA) was performed to analyze inflammatory biomarkers and adipokines.

Results: Distribution frequency of TNF-α -857CT (OR=3.316, 95% CI=1.092–8.304, p=0.025) in women with GDM pregnancies were obviously higher than that in women with healthy pregnancies. Women with GDM were of older maternal age, had higher BMI, were more nulliparous, and had T2DM and GDM history, compared to women with healthy pregnancies (p<0.05). Inflammatory biomarkers in serum (hs-CRP, IL-6, IL-8, IL-6/IL-10 ratio) and placental (NF-κB, IL-6, IL-8, IL-6/IL-10 ratio, IL-1β, TNF-α) were significantly different (p<0.05) between women with GDM and women with healthy pregnancies. Differences were found for serum FBG, FINS, HOMA-IR, and HOMA-β, and placental IRS-1, IRS-2, leptin, adiponectin, visfatin, RBP-4, chemerin, nesfatin-1, FATP-4, EL, LPL, FABP-1, FABP-3, FABP-4, and FABP-5.

Conclusions: TNF-α -857C/T SNP, hs-CRP, IL-6, IL-8, and IL-6/IL-10 were associated with GDM in women from Inner Mongolia, as was serious inflammation and disordered lipid and glucose metabolisms.

MeSH Keywords: Diabetes, Gestational • Inflammation • Metabolism • Polymorphism, Single Nucleotide

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Background

Gestational diabetes mellitus (GDM) is a common endocrine and metabolic disease with different degrees of glucose and lipid metabolic disorders during pregnancy, causing danger to the mothers and the perinatal infants, and can even be life-threatening [1,2]. However, the pathogenesis of GDM is still unclear, and might be related to hormone level changes, insulin resistance (IR), obesity, a chronic inflammatory reaction, oxidative stress, genetics, insulin-secretion deficiency, immunity and so on [3].

Recently attention has focused on the relationship between inflammatory reaction and IR, indicating that inflammation is a critical factor in promoting IR. During pregnancy, the maternal tissues become insensitive to insulin resulting from changes due to placental lactogen and other hormones. For women with GDM, IR results in maternal hyperglycemia and stimulates fetal hyperinsulinemia [4]. IR and reduced secretory function of β cell are important pathological features of GDM.

After giving birth, GDM in women disappears immediately. Risk factors, such as increased maternal age, previous occurrence of GDM, and family history of diabetes, are helpful for diagnosis of GDM [5].

In China, a universal screening program for GDM in women living in the six urban districts of Tianjin was carried out, with the results showing that the GDM prevalence was as low as 2.3% in 1998 [6]. However, there is scant research on the prevalence of GDM in the Chinese population, and in particular Inner Mongolia.

In our study, we analyzed the expressions of IL-65–72C/G and TNF-α -857C/T SNPs in GDM patients from Inner Mongolia, as well as their relationship with glucose and lipid metabolism, in order to inform clinical diagnosis and prevention of GDM with information on related molecular mechanisms.

Material and Methods

Patients

Data were collected from 140 women with GDM pregnancies and 140 women with healthy pregnancies, who delivered in the Obstetric Department of Huhhot First Hospital from June of 2013 to June of 2015. All the women lived in Inner Mongolia (China) for more than 20 years, with routine ultrasonic examination and prenatal care. Informed consent was obtained from each participant and approved by the medical ethics committee of Huhhot First Hospital and Zhongnan Hospital of Wuhan University.

GDM pregnancies were diagnosed according to the criteria of Endocrine Society Clinical Practice Guideline [7] and American College of Obstetricians and Gynecologists (ACOG). The pregnant women, with gestational weeks of 24~28, were screened for GDM by one hour 50 g glucose challenge test (GCT). When the fasting glucose was higher than 92 mg/d/L but lower than 126 mg/d/L, GDM was diagnosed at any gestational age. Those pregnant women in early pregnancy, not meeting the diagnostic criteria for GDM, were asked to have oral glucose tolerance test (OGTT) from the 24th and 28th gestational weeks [7].

Genomic DNA extraction and polymerase chain reaction (PCR)

We collected 10 mL peripheral blood at 9: 00 in the morning, which was then store at –28°C for subsequent experiments. A sample of 2 mL of the blood was used for SNPs analysis. Genomic DNA in each sample was extracted and purified with genomic DNA extraction kit (Thermo Fisher Scientific, Inc., Shanghai, China) according to the kit specification, and then stored at –78°C until PCR amplification.

PCR primers were designed with Sequenom Assay Desigh 3.0 software and synthesized by Thermo Fisher. Primers are shown in Table 1. DNA samples were diluted quantitatively and then prepared for amplification. PCR amplification system is shown in Table 2, with the process in Table 3. After amplification, 0.3 U alkaline phosphatase was added to remove the remaining

Table 1. Primers for amplification on IL-6–572C/G and TNF-α -857C/T.

Gene *loci*	Primers	Endonuclease
IL-6–572C/G	F: 5'-GGA GAC GCC TTG AAG TAA CTG C-3'	AseI
	R: 5'-GAG TTT CCT CTG ACT CCA TCG CAG-3'	
TNF-α -857C/T	F: 5'-AAG TCG AGT ATG ATG GGG ACC CCC CAT TAA-3'	MbiI
	R: 5'-AAG CTC TCA CTT CTC AGG GCC CCA GT-3'	

IL-6 – interleukin-6; TNF-α – tumor necrosis factor-α; F – forward primer; R – reverse primer.

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Table 2. PCR amplification system for IL-6–572C/G and TNF-α -857C/T.

	IL-6–572C/G (μL)	TNF-α -857C/T (μL)
DNA template	5	2
Primers	2	F: 1
	2	R: 1
dNTP(1×)	4	2
Taq polymerase (1×)	0.4	0.5
PCR buffer (1×)	5	5
Total	**50**	**50**

PCR – polymerase chain reaction; IL-6 – interleukin-6; TNF-α – tumor necrosis factor-α; dNTP – deoxyribonucleotide triphosphate.

deoxyribonucleotide triphosphate (dNTP). Finally the PCR products were spotted into SpectroCHIP (Sequenom) with the automatic instrument. MALDI-TOF-MS (SpectroREADER, Sequenom) was used to detect the chips.

Metabolic and inflammatory biomarkers

High sensitivity C-reactive protein (hs-CRP), fasting blood glucose (FBG), and fasting blood insulin (FINS) were detected by automatic biochemical analyzer (BioTek Instrument, Inc., Beijing, China). Homeostasis Model Assessment (HOMA) formula was used for calculating IR index (HOMA-IR) and islet β cell function index (HOMA-β). Serum Interleukin-1β (IL-1β), IL-6, IL-8, IL-10, and tumor necrosis factor-α (TNF-α) were determined by Bio-Plex 200 Systems with Bio-Plex ProTM Human Cytokine Assay kits (Bio-Rad Laboratories, Inc., Shanghai, China).

Placental tissues

Fresh placental tissues were collected after delivery and fixed in 10% formalin (Solarbio Technology Co., Ltd., Beijing. China), and then finely ground with RIPA buffer (Thermo). Tissue residues were discarded after centrifuging. Tissue suspension was used for detecting placental indexes by enzyme linked immunosorbent assay (ELISA), including nuclear factor-κB (NF-κB), TNF-α, IL-1β, IL-6, IL-8, IL-10, leptin, adiponectin, visfatin, retinol-binding protein-4 (RBP-4), chemerin, nesfatin-1, insulin receptor substrate-1 (IRS-1), IRS-2, fatty acid transport protein-4 (FATP-4), endothelial lipase (EL), low density lipoprotein (LPL), fatty acid binding protein-1 (FABP-1), FABP-3, FABP-4 and FABP-5. ELISA kits were obtained from TaKaRa Biotechnology Co., Ltd. (Dalian, China), Beijing Wantai Biological Pharmacy Enterprise Co., Ltd. (China) and Shanghai Kehua Bio-Engineering Co., Ltd. (China).

Statistical analysis

All data were analyzed by SPSS 19.0 and shown as mean±standard deviations (X ±SD). Hardy-Weinberg equilibrium (HWE) was used

Table 3. PCR amplification conditions for IL-6–572C/G and TNF-α -857C/T.

Step	Temperature (°C)	Time	Cycle number
IL-6–572C/G			
Predegeneration	94°C	5 min	
Degeneration	94°C	45 s	
Annealing	61°C	45 s	30 cycles
Extension	72°C	45 s	
Extension terminal	72°C	10 min	1 cycle
Storage	4°C	→∞	
TNF-α -857C/T			
Predegeneration	94°C	15 min	
Degeneration	94°C	30 s	
Annealing	57°C	30 s	35 cycles
Extension	72°C	30 s	
Extension terminal	72°C	3 min	1 cycle
Storage	4°C	→∞	

PCR – polymerase chain reaction; IL-6 – interleukin-6; TNF-α – tumor necrosis factor-α.

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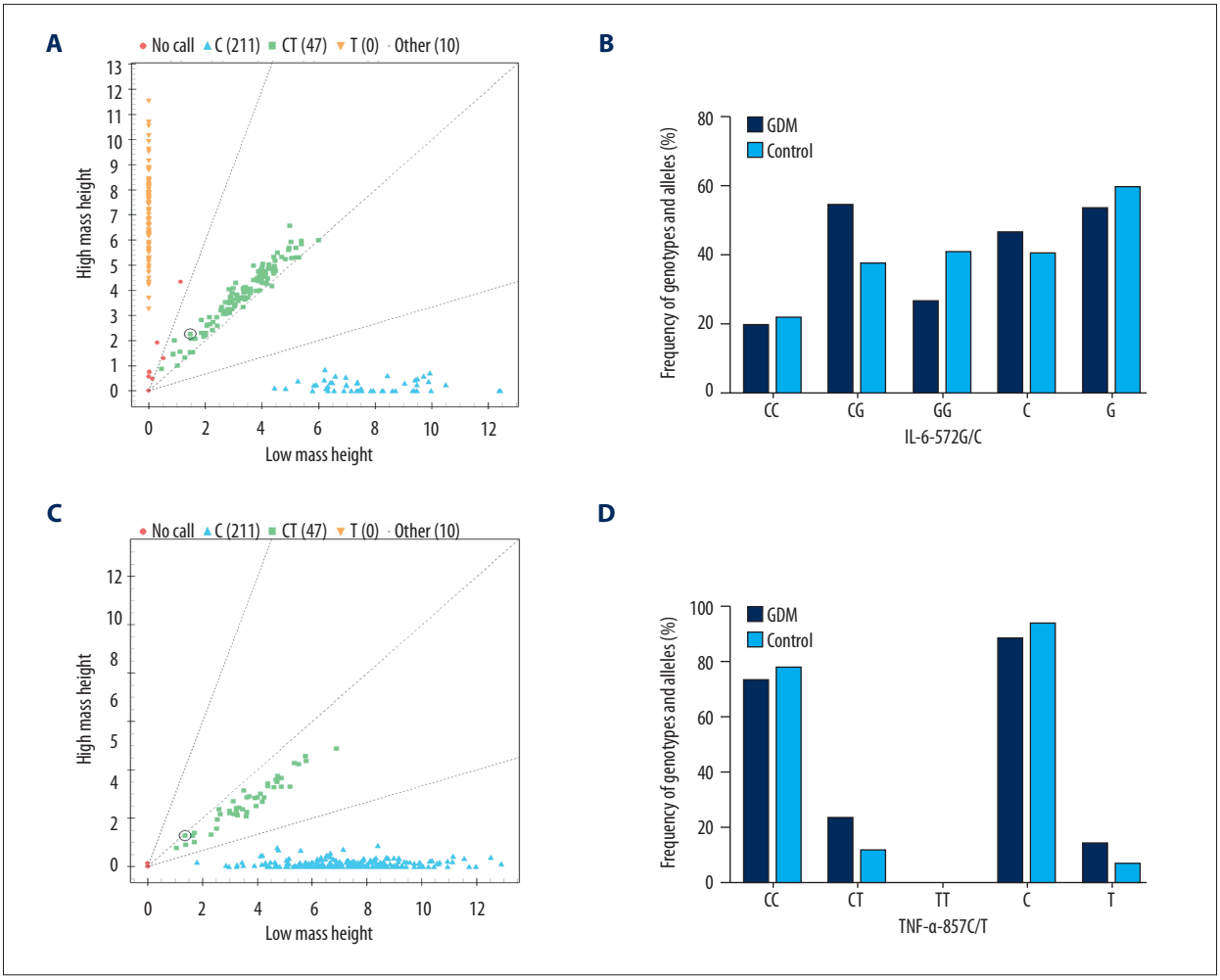

Figure 1. Comparisons of distribution frequencies of genotypes and alleles on IL-65–72C/G and TNF-r -857C/T in pregnancies with or without GDM. (**A**) Mass spectrometry for IL-65–72C/G; (**B**) distribution frequencies for IL-65–72C/G; (**C**) mass spectrometry for TNF-α -857C/T; (**D**) distribution frequencies for TNF-e -857C/T). GDM – gestational diabetes mellitus; IL-6 – interleukin-6; TNF-α – tumor necrosis factor-α.

for estimating allele frequencies. Comparisons of genotypes and allele frequencies between the patients and volunteers were analyzed by chi-square test. Binary logistic regression analysis was used for evaluating the association between SNPs of IL-6, TNF-α genotype, and allele frequencies with GDM. The characteristics were compared with Fisher's exact test. T-test was used for comparisons on continuous variables. Logistic regression was used for the association of inflammatory biomarkers in serum with GDM. A p value <0.05 was considered significant.

Results

IL-65–72C/G and TNF-α -857C/T

Mass spectrometry of IL-65–72C/G and TNF-α -857C/T genotypes are shown in Figure 1 and Table 4. Both allele frequencies of IL-65–72C/G (χ^2=0.6712, p>0.05) and TNF-α -857C/T (χ^2=0.3909, p>0.05) fit the Hardy-Weinberg genetic equilibrium.

As shown in Figure 1A and 1B, there were 20 samples of GDM pregnancies and healthy pregnancies without results. The distribution frequencies of IL-65–72C/G in GDM pregnancies were: CC (19.29%), CG (54.29%), and GG (26.43%). The allele frequencies were C (46.43%) and G (53.57%), respectively. In healthy pregnancies, IL-65–72C/G frequencies were CC (21.67%), CG (37.50%), and GG (40.83%), and the allele frequencies were C (40.42%) and G (59.58%). There was no significant difference on genotypes or allele frequencies of IL-65–72C/G between GDM and healthy pregnancies.

As shown in Figure 1C and 1D, there were six samples of GDM pregnancies and 16 of healthy pregnancies without results. The distribution frequencies of TNF-α -857C/T in GDM were

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Table 4. Distribution of IL-6–572C/G and TNF-α -857C/T polymorphism in GDM and healthy pregnancies.

Genotypes	GDM (n, %)	Volunteers (n, %)	OR	95% CI	P value
IL-6–572C/G					
CC	27 (19.29)	26 (21.67)	1.025	0.462–2.961	0.844
CG	76 (54.29)	45 (37.50)	2.836	1.105–7.368	0.031
GG	37 (26.43)	49 (40.83)	1.097	1.433–6.995	0.046
C	130 (46.43)	97 (40.42)	–	–	0.036
G	150 (53.57)	143 (59.58)	–	–	–
TNF-α -857C/T					
CC	102 (72.86)	109 (77.86)	1.265	0.328–3.570	0.783
CT	32 (22.86)	15 (10.71)	3.316	1.092–8.304	0.025
TT	0 (0)	0 (0)	–	–	–
C	236 (88.06)	233 (93.95)	–	–	0.028
T	32 (11.94)	15 (6.05)	–	–	–

IL-6 – interluekin-6; GDM – gestational diabetes mellitus; OR – odd ratio; CI – confidence interval.

Table 5. Comparisons on clinical characteristics, serum metabolic indexes and inflammatory biomarkers of the pregnancies with or without GDM (X ±SD) (N=120).

	GDM (n=60)	Control (n=60)	P
Maternal age (years)	29.13±3.65	25.06±5.94	0.001*
Pre-pregnancy BMI (kg/m^2)	38.68±9.50	28.74±7.26	0.000*
Nulliparous	10 (16.67%)	28 (46.67%)	0.019*
Gestational age (weeks)	21.3±4.05	21.8±4.30	0.811
Family history of T2DM	34 (56.67%)	13 (21.67%)	0.008*
History of GDM	8 (113.33%)	1 (1.67%)	0.042*
FBG (mmol/L)	8.03±1.57	4.20±1.62	0.001*
FINS (mIU/L)	3.15±1.08	1.92±1.54	0.001*
HOMA-IR	4.89±1.40	2.16±1.51	0.001*
HOMA-β	75.32±10.64	157.49±18.85	0.000*
hs-CRP (mg/L)	8.38±1.67	4.60±1.04	0.000*
IL-6 (ng/L)	5.85±1.41	3.91±1.66	0.002*
IL-8 (ng/L)	2.17±0.45	0.74±0.12	0.000*
IL-10 (ng/L)	1.86±0.52	2.03±0.39	0.502
IL-6/IL-10 ratio	3.15±0.73	1.93±0.46	0.000*
IL-1β (ng/L)	0.89±0.64	0.92±0.51	0.847
TNF-α (ng/L)	8.60±1.08	7.21±1.19	0.320

GDM – gestational diabetes mellitus; BMI – body mass index; T2DM – type 2 diabetes mellitus; hs-CRP – high sensitivity C-reactive protein; IL – interleukin; TNF-α – tumor necrosis factor-α; FBG – fasting blood glucose; FINS – fasting blood insulin; HOMA – homeostasis model assessment; IR – insulin resistance; X±SD – mean ± standard deviations; * P<0.05.

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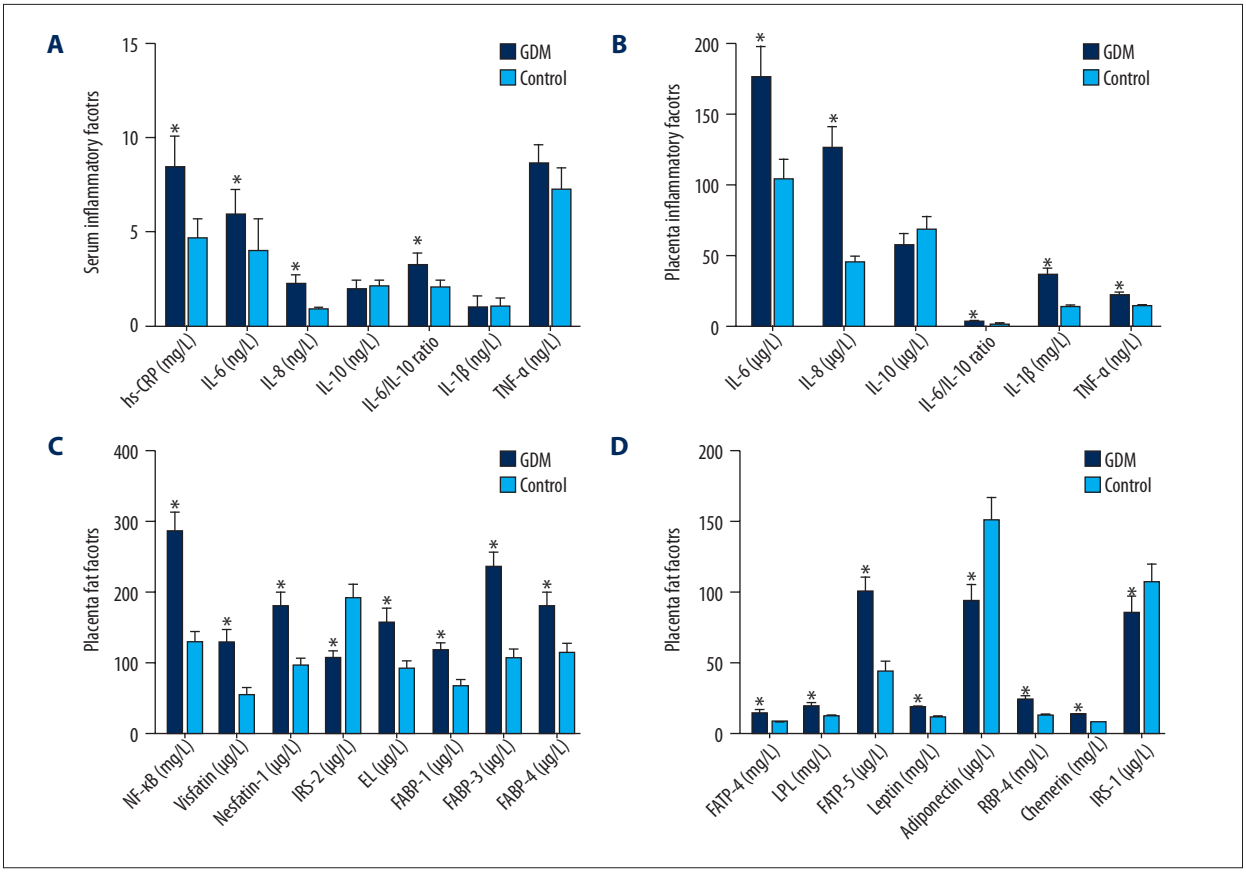

Figure 2. Comparisons on serum and/or placental inflammatory biomarkers and/or adipokines of pregnancies with or without GDM. (**A**) Serum inflammatory factors; (**B**) placental inflammatory factors and adipokines, * $p<0.05$). GDM – gestational diabetes mellitus; hs-CRP – high sensitivity C-reactive protein; IL – interleukin; TNF-α – tumor necrosis factor-α; NF-κB – nuclear factor-κB; RBP – retinol-binding protein; irs-1 – insulin receptor substrate-1; FATP – fatty acid transport protein; EL – endothelial lipase; LPL – low density lipoprotein; FABP – fatty acid binding protein.

CC (72.86%) and CT (22.86%), and allele frequencies were C (88.06%) and T (13.56%). In healthy pregnancies, the distribution frequencies of TNF-α -857C/T were CC (77.86%) and CT (10.71%), and the allele frequencies were C (93.95%) and T (6.05%), respectively. The differences of the two genotypes frequencies were statistically significant between GDM and healthy pregnancies (chi-square=5.84, $p<0.05$), as well as the allele frequencies (chi-square=6.97, $p<0.05$). The distribution frequency of CT in GDM pregnancies was significantly higher than in healthy pregnancies (OR=3.316, 95% CI=1.092–8.304, $p=0.025$), which showed that the risk of developing GDM for patients carrying CT genotype.

Clinical characteristics, serum metabolic and inflammatory biomarkers

Table 5 and Figure 2A show the results of the comparisons of clinical characteristics, serum metabolic biomarkers, and inflammatory biomarkers of the pregnancies with or without GDM. There were significant differences ($p<0.05$) between the GDM pregnancies and the healthy pregnancies in clinical characteristics, including maternal age (29.13±3.65 versus 25.06±5.94 years), pre-pregnancy body mass index (BMI) (38.68±9.50 versus 28.74±7.26), nulliparous women (10 women versus 28 women), family history of type 2 diabetes mellitus (T2DM) (34 women versus 13 women), and history of GDM (8 women versus 1 woman), but not for gestational age. Additionally, serum inflammatory biomarkers were also significant different ($p<0.05$) between GDM pregnancies and healthy pregnancies, including hs-CRP (8.38±1.67 versus 4.60±1.04 mg/L), IL-6 (5.85±1.41 versus 3.91±1.66 ng/L), IL-8 (2.17±0.45 versus 0.74±0.12 ng/L), and IL-6/IL-10 ratio (3.15±0.73 versus 1.93±0.46), while excluding IL-10, IL-1β, and TNF-α.

Blood glucose metabolic indexes in serum were significantly different between GDM pregnancies and healthy pregnancies, including FBG (8.03±1.57 versus 4.20±1.62 mmol/L), FINS (3.15±1.08 versus 1.92±1.54 mIU/L), HOMA-IR (4.89±1.40 versus 2.16±1.51) and HOMA-β (75.32±10.64 versus 157.49±18.85), respectively ($p<0.05$).

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Table 6. Association of serum inflammatory biomarkers with GDM (X ±SD).

Inflammatory biomarkers	X ±SD	OR	95% CI	P
Log (CRP)	1.25±1.03	9.52	2.76, 33.10	0.001*
Log (IL-6)	1.29±0.67	4.66	1.53, 14.59	0.008*
Log (IL-8)	1.03±0.42	2.58	1.27, 10.08	0.009*
Log (IL-10)	0.61±0.32	0.46	1.19, 2.43	0.408
IL-6/IL-10 ratio	2.14±1.08	1.75	1.22, 2.68	0.003*
Log (IL-1β)	–0.42±0.57	1.04	0.37, 2.44	0.995
Log (TNF-α)	1.70±0.66	1.02	0.35, 2.73	0.452

GDM – gestational diabetes mellitus; CRP – C-reactive protein; IL – interleukin; TNF-α – tumor necrosis factor-α; X ±SD – mean ± standard deviations; * P<0.05.

Association of serum inflammatory biomarkers with GDM

As shown in Table 6, some of the serum inflammatory biomarkers were obviously associated with GDM, including CRP (0R=9.52, 95% CI (2.76, 33.10), *p*=0.001), IL-6 (OR=4.66, 95% CI (1.53, 14.59), *p*=0.008), IL-8 (OR=2.58, 95% CI (1.27, 10.08), *p*=0.009) and IL-6/IL-10 ratio (OR=1.75, 95% CI (1.22, 2.68), *p*=0.003).

Inflammatory biomarkers and adipokines in placentas

After delivery, inflammatory biomarkers and adipokines in placentas were detected and compared. The results are shown in Table 7.

In Figure 2B, except for IL-10, all the inflammatory biomarkers in placentas had significant differences ($p<0.05$) between GDM pregnancies and healthy pregnancies, including NF-κB (283.56±31.32 versus 125.69±17.38 μg/L), IL-6 (177.09±22.68 versus 103.85±15.6 μg/L), IL-8 (126.43±16.02 versus 45.70±4.95 μg/L), IL-6/IL-10 ratio (3.09±1.40 versus 1.51±1.06), IL-1β (36.42±5.10 versus 13.48±2.06 mg/L) and TNF-α (21.61±3.21 versus 13.82±2.02 mg/L). IRS-1 (83.94±13.22 versus 106.01±14.30 μg/L) and IRS-2 (104.28±10.95 versus 190.11±20.58 μg/L) in placentas were significantly different ($p<0.05$) between GDM pregnancies and healthy pregnancies. Lipid metabolic indexes all had significant differences ($p<0.05$) between GDM pregnancies and healthy pregnancies as follows: leptin (16.36±2.03 versus 9.17±1.88 mg/L), adiponectin (92.58±13.20 versus 150.14±17.21 μg/L), visfatin (127.39±18.68 versus 52.28±9.05 μg/L), RBP-4 (22.46±3.01 versus 10.72±2.05 mg/L), chemerin (11.20±1.17 versus 5.76±0.03 mg/L), nesfatin-1 (178.32±20.77 versus 93.59±11.64 μg/L), FATP-4 (13.06±2.90 versus 6.50±0.75 mg/L), EL (154.91±20.16 versus 89.04±10.37 μg/L), LPL (17.14±3.68 versus 10.32±1.07 mg/L), FABP-1 (115.60±12.44 versus 63.38±9.83 μg/L), FABP-3 (235.17±20.84 versus 105.26±13.02 μg/L), FABP-4 (178.37±20.98 versus 112.64±14.02 μg/L) and FABP-5 (99.40±11.37 versus 42.31±8.29 μg/L).

Discussion

In our study, we firstly detected the SNPs of IL-65–72C/G and TNF-α -857C/T in GDM and healthy pregnancies. The results showed that the differences in the CC and CT genotypes of TNF-α -857C/T were statistically significant between GDM and healthy pregnancies, while IL-65–72C/G was without significance. The distribution frequency of TNF-α -857CT in GDM pregnancies was higher than that in healthy pregnancies, which indicated a risk of developing GDM for patients carrying TNF-α -857CT, and T allele was the risk factor.

According to previous studies, IL-65–72C/G is associated to obesity [8,9], colon cancer [10], and T2DM [11]; as is TNF-α -857C/T [7,8], but no research has focused on their relationship with GDM. In our study, we did not find an association of IL-65–72C/G with GDM pregnancies. Therefore, we presumed that women carrying TNF-α -857CT in Inner Mongolia were at higher risk for GDM than those without.

In addition, by detecting serum factors we found that hs-CRP, IL-6, IL-8, and IL-6/IL-10 were significantly associated with GDM. The inflammatory cytokines TNF-α [12], IL-6 [13,14], hs-CRP [14,15], IL-1 β [16,17], and IL-8 [18] have mostly been found to be increased in GDM pregnancies compared to healthy pregnancies. However, in our study, TNF-α and IL-1β was found to be abnormal in GDM pregnancies, but not significantly. This difference might be due to differences in pregnancies within different population, indicating that for GDM women in Inner Mongolia, abnormal levels of hs-CRP, IL-6, IL-8, and IL-6/IL-10 ratio may be diagnostic biomarkers.

In women with GDM, the placenta may adapt to the inflammatory environment and exhibit morphological changes [19–21]. In our study, we collected the placentas from GDM and healthy pregnancies. By detecting the metabolic indexes, we found that the inflammatory biomarkers, including NF-κB, IL-6, IL-8,

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Table 7. Inflammatory biomarkers and fat factors in placentas of the pregnancies with or without GDM (X ±SD) (N=120).

	GDM (n=60)	Control (n=60)	P
NF-κB (μg/L)	283.56±31.32	125.69±17.38	0.000*
IL-6 (μg/L)	177.09±22.68	103.85±15.6	0.002*
IL-8 (μg/L)	126.43±16.02	45.70±4.95	0.000*
IL-10 (μg/L)	57.32±9.44	68.65±9.33	0.639
IL-6/IL-10 ratio	3.09±1.40	1.51±1.06	0.000*
IL-1β (mg/L)	36.42±5.10	13.48±2.06	0.000*
TNF-α (mg/L)	21.61±3.21	13.82±2.02	0.006*
Leptin (mg/L)	16.36±2.03	9.17±1.88	0.000*
Adiponectin (μg/L)	92.58±13.20	150.14±17.21	0.000*
Visfatin (μg/L)	127.39±18.68	52.28±9.05	0.000*
RBP-4 (mg/L)	22.46±3.01	10.72±2.05	0.002*
Chemerin (mg/L)	11.20±1.17	5.76±0.03	0.005*
Nesfatin-1 (μg/L)	178.32±20.77	93.59±11.64	0.000*
IRS-1 (μg/L)	83.94±13.22	106.01±14.30	0.010*
IRS-2 (μg/L)	104.28±10.95	190.11±20.58	0.000*
FATP-4 (mg/L)	13.06±2.90	6.50±0.75	0.000*
EL (μg/L)	154.91±20.16	89.04±10.37	0.000*
LPL (mg/L)	17.14±3.68	10.32±1.07	0.003*
FABP-1 (μg/L)	115.60±12.44	63.38±9.83	0.000*
FABP-3 (μg/L)	235.17±20.84	105.26±13.02	0.000*
FABP-4 (μg/L)	178.37±20.98	112.64±14.02	0.000*
FABP-5 (μg/L)	99.40±11.37	42.31±8.29	0.000*

GDM – gestational diabetes mellitus; NF-κB – nuclear factor-κB; IL – interleukin; TNF-α – tumor necrosis factor-α; RBP – retnol-binding protein; irs-1 – insulin receptor substrate-1; FATP – fatty acid transport protein; EL – endothelial lipase; LPL – low density lipoprotein; FABP – fatty acid binding protein; X ±SD – mean ± standard deviations; * P<0.05.

IL6/IL-10 ratio, IL-1β, and TNF-α, had significant differences between GDM and healthy pregnancies, as well as the adipokines, including leptin, adiponectin, visfatin, RBP-4, chemerin and nesfatin-1, IRS-1, IRS-2, FATP-4, EL, LPL, FABP-1, FABP-3, FABP-4, and FABP-5. This phenomenon showed that the placentas of GDM women were in an inflammatory environment, and that the increased inflammatory mediators might promote intraplacental inflammatory cascades by specific gene transcription or translation. Additionally, the adipose tissue also generated numbers of adipokines to promote the production of inflammatory cytokines, which aggravated the GDM status.

Otherwise, we found that both FBG and FINS in serum obviously increased, and the HOMA-IR and HOMA-β were significantly different between GDM and healthy pregnancies. Considering the decreased IRS-1 and IRS-2 in placentas of GDM, we confirmed that glucose metabolism was disordered in the whole body, a finding which was in accorded with previous studies [22,23], as was lipid metabolism [24,25].

However, which cytokines were regulated and how deregulation led to the GDM progression, is still unclear. In the future, there should be further investigations using larger sample sizes and collecting data from pregnant women in different regions.

Conclusions

Our study confirmed that TNF-α -857C/T SNPs and inflammation were associated with GDM, and glucose and lipid metabolisms were disordered in GDM.

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Statement

No conflict in this study declared by all authors. There is no foundation supporting this work.

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