Clin. Lab. 2021;67:XXX-XXX

ORIGINAL ARTICLE

Correlation of Homocysteine, AHSG, CRP with Insulin Resistance, 25-(OH)2-VitD, Blood Lipids in Gestational Diabetes Patients

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SUMMARY

Background: **To investigate the correlations of serum homocysteine (Hcy), α2-Heremans-Schmid glycoprotein (AHSG), and C-reactive protein (CRP) with insulin resistance (IR), 25-hydroxyvitamin D (25-OH-VD), and blood lipids in patients with gestational diabetes mellitus (GDM) by detecting their levels.**

Methods: **A total of 72 GDM patients (GDM group) and 72 healthy pregnant women (control group) delivered in our hospital from February 2017 to January 2019 were randomly selected. The basic data, somatological parameters [height, weight, body mass index (BMI), waist circumference, hip circumference, waist-to-hip ratio (WHR), blood pressure, and body fat content], and biochemical indexes (glucose metabolism indexes, lipid metabolism indexes, Hcy, AHSG, CRP, and 25-OH-VD) were compared between the two groups. Additionally, Pearson's correlation analysis was employed to analyze the correlations among indicators.**

Results: **In comparison with the control group, the GDM group had a higher average rate of family history of DM ($p < 0.05$), larger waist circumference and WHR, and higher body fat content ($p < 0.05$). Besides, the fasting plasma glucose (FPG), 1-hour plasma glucose (1hPG) and 2-hour plasma glucose (2hPG), glycosylated hemoglobin (HbA1c), fasting insulin (FINS), homeostasis model assessment (HOMA)-IR, triglyceride (TG), total cholesterol (TC), and low density lipoprotein cholesterol (LDL-C) were higher in the GDM group than those in the control group ($p < 0.05$), while the high density lipoprotein cholesterol (HDL-C) was lower in the GDM group than that in the control group ($p < 0.05$). Compared with those in the control group, the serum Hcy, AHSG, and CRP levels rose, while the serum 25-OH-VD level declined in the GDM group ($p < 0.05$). The results of Pearson's correlation analysis revealed that HOMA-IR had positive correlations with FPG, FINS, TC, TG, Hcy, AHSG, and CRP ($r = 0.591, 0.825, 0.312, 0.234, 0.458, 0.647, 0.487$, $p < 0.05$) and negative correlation with 25-OH-VD ($r = -0.323$, $p < 0.05$). CRP was positively correlated with HOMA-IR, TC, and AHSG ($r = 0.485, 0.331, 0.226$, $p < 0.05$), negatively associated with 25-OH-VD ($r = -0.443$, $p < 0.05$), and had no correlation to TG and Hcy ($r = 0.019, 0.058$, $p > 0.05$). AHSG displayed positive correlations with HOMA-IR, TC, TG, and CRP ($r = 0.647, 0.321, 0.314, 0.226$, $p < 0.05$) and no association with Hcy and 25-OH-VD ($r = 0.058, -0.034$, $p > 0.05$).**

Conclusions: **GDM patients have increased serum Hcy, AHSG, and CRP levels and a decreased serum 25-OH-VD level, indicating that serum Hcy, AHSG, CRP, and 25-OH-VD are correlated with glucose and lipid metabolism disorders in GDM patients.**

(Clin. Lab. 2021;67:xx-xx. DOI: 10.7754/Clin.Lab.2020.200609)

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Manuscript accepted July 10, 2020

KEY WORDS

GDM, correlation, IR

INTRODUCTION

Gestational diabetes mellitus (GDM), a common pregnancy complication in obstetrics, refers to abnormal glucose metabolism first discovered or occurring during pregnancy. GDM is a factor of high-risk pregnancy. In the short term, it will increase the incidence rates of pregnancy complications and adverse pregnancy outcomes [1]. In the long term, pregnant women and their offspring will be more prone to develop type 2 DM (T2DM), hypertension, and obesity [2], with severely affected health of fetuses, neonates, and gravidas.

Studies have confirmed that the main pathogenesis of GDM is related to insulin resistance (IR) [3,4]. Another study showed that subclinical inflammatory reactions are also involved in the pathogenesis of GDM [5]. Increased inflammatory factor C-reactive protein (CRP) is associated with hyperglycemia, which is considered to be an independent risk factor for IR and diabetes mellitus (DM) [6,7]. Research found that glycoprotein-α2–Heremans-Schmid glycoprotein (AHSG) produced by the liver affects glucose metabolism by repressing the phosphorylation and tyrosinase activity of insulin receptors [8]. Studies have manifested that homocysteine (Hcy) is significantly elevated in the serum of GDM patients, and it is also closely related to T2DM and cardiovascular diseases [9-11]. Studies have found that patients with diabetes and metabolic syndrome have varying degrees of vitamin D deficiency [12,13], and serum 25-hydroxyvitamin D (25-OH-VD) is the best indicator to measure the nutritional status of the body's vitamin D. Therefore, in this article, levels of the serum Hcy, AHSG, CRP, IR, 25-OH-VD, and blood lipid were measured in GDM patients. The correlations between the indicators were explored, so as to understand the influencing factors of GDM and provide evidence-based medical evidence to reduce the incidence rate of GDM and prevent and control GDM.

MATERIALS AND METHODS

Clinical data

A total of 72 GDM patients (GDM group) and 72 healthy pregnant women (control group) were randomly selected in our hospital from February 2017 to January 2019. At 24 - 28 weeks of pregnancy, all subjects were subjected to an oral glucose tolerance test (OGTT), with reference to the 2010 international diagnostic criteria for GDM [14]. For OGTT, the normal values of fasting plasma glucose (FPG), 1-hour plasma glucose (1hPG) and 2-hour plasma glucose (2hPG) values are < 5.1, 10.0, and 8.5 mmol/L, respectively. Blood glucose greater than or equal to the above three values indicates GDM. Inclusion criteria: (1) women with a naturally conceived singleton pregnancy, (2) those aged 20 - 45 years old, and (3) those undergoing OGTT at 24 - 28 weeks of pregnancy. Exclusion criteria: patients with previous history of DM, taking folic acid and/or B vitamins within the past 6 weeks, or suffering from severe heart, liver or kidney diseases. All patients enrolled were informed of the study and signed the informed consent. This study was approved by the Ethics Committee of our hospital.

Observation indexes and detection methods

General clinical data

General clinical data such as patient name, age, menstrual history, marriage and childbearing history, gestational age, family history and medication history were recorded.

Somatological parameters

Height (without shoes and hat) and weight of subjects wearing light clothing and with an empty stomach were measured. Body mass index (BMI) = weight/height^2 (kg/m^2). The waist circumference (between the lowermost rib and the crista iliaca) and hip circumference (at the widest portion of the hip) were measured using a soft ruler with the subjects standing upright.

Detection of biochemical indicators

At 24 - 28 weeks of pregnancy, all subjects were subjected to OGTT. After fasting for 10 hours, blood samples were collected from the anterior cubital veins the next morning for detection. Fluorescence polarization immunoassay was adopted for the determination of plasma total homocysteine (tHcy), and AHSG was detected via enzyme-linked immunoassay. Immunoturbidimetry was employed for high sensitivity CRP (hs-CRP) determination, and 25-OH-VD was measured by electroluminescence. Micro-column assay was conducted for measurement of glycosylated hemoglobin (HbA1c), glucose oxidase assay for FPG, 1hPG and 2hPG, and radioimmunoassay for fasting insulin (FINS). IR was assessed using homeostasis model assessment (HOMA) [15]. HOMA-IR = FPG x FINS/22.5. Enzyme method was used to measure serum triglyceride (TG), total cholesterol (TC), low-density lipoprotein cholesterol (LDL-C), and high-density lipoprotein cholesterol (HDL-C). All assays were performed by a person specially assigned in strict accordance with the instructions.

Data statistics and analysis

Statistical Product and Service Solutions (SPSS) 23.0 software (IBM SPSS Statistics for Windows; IBM Corp, Armonk, NY, USA) was used to record data. Measurement data were expressed as mean ± standard deviation ($\overline{x} \pm s$), and subjected to a *t*-test. Enumeration data were expressed as n, and χ^2 test was adopted. Pearson's correlation analysis was employed. $p < 0.05$ suggested that the difference was statistically significant.

RESULTS

Comparison of basic data of patients between the two groups

The results of basic situation comparison revealed that the GDM group had a raised rate of family history of DM in comparison with the control group, and the difference was statistically significant ($p < 0.05$). There were no statistically significant differences in the mean age, gestational age, gravidity, and family history of cardiovascular diseases between the two groups ($p > 0.05$) (Table 1).

Comparisons of somatological parameters between two groups of patients

The waist circumference, waist-to-hip ratio (WHR) and body fat were larger in the GDM group than those in the control group, showing statistical differences ($p < 0.05$), while no statistically significant differences were found in the weight before pregnancy, height, BMI before pregnancy, and hip circumference between the two groups of patients ($p > 0.05$) (Table 2, Figure 1).

Comparison of glucose and lipid metabolism indexes between the two groups of patients

Compared with those in control group, the FPG, 1hPG, 2hPG, HbA1c, FINS, and HOMA-IR were increased in the GDM group, and the differences were statistically significant ($p < 0.05$). In comparison with the control group, the GDM group exhibited elevated levels of TC, TG, and LDL-C and a reduced HDL-C level, and the differences were of statistical significance ($p < 0.05$) (Table 3, Figure 2).

Comparison of serum Hcy, AHSG, CRP, and 25-OH-VD of patients between the two groups

The serum Hcy, AHSG, and CRP levels were clearly higher in the GDM group than those in the control group [(18.12 ± 5.90) vs. (9.54 ± 3.57), (250.56 ± 23.12) vs. (163.84 ± 17.39), and (9.03 ± 3.17) vs. (2.41 ± 0.62), respectively], showing statistical differences ($p < 0.05$), while the 25-OH-VD level was lower in the GDM group than that in the control group [(15.37 ± 4.45) vs. (19.38 ± 4.72)], with a statistical difference ($p < 0.05$) (Table 4, Figure 3).

Correlation analysis of HOMA-IR with various serum indicators

It was uncovered through Pearson's correlation analysis that HOMA-IR was positively correlated with FPG, FINS, TC, TG, Hcy, AHSG, and CRP ($r = 0.591, 0.825, 0.312, 0.234, 0.458, 0.647, 0.487$, $p < 0.05$) and negatively associated with 25-OH-VD ($r = -0.323$, $p < 0.05$) (Table 5).

Correlation analysis of CRP with various serum indexes

The results of Pearson's correlation analysis showed that CRP displayed positive correlations with HOMA-IR, TC, and AHSG ($r = 0.485, 0.331, 0.226$, $p < 0.05$), a negative correlation to 25-OH-VD ($r = -0.443$, $p < 0.05$), and no obvious correlation to TG and Hcy ($r = 0.019, 0.058$, $p > 0.05$) (Table 6).

Correlation analysis of AHSG with various serum indicators

According to Pearson's correlation analysis, AHSG was positively associated with HOMA-IR, TC, TG, and CRP ($r = 0.647, 0.321, 0.314, 0.226$, $p < 0.05$) and had no evident correlation to Hcy and 25-OH-VD ($r = 0.058, -0.034$, $p > 0.05$) (Table 7).

DISCUSSION

A study revealed that the elevated level of maternal serum Hcy during pregnancy is closely related to the occurrence of multiple abnormal pregnancy outcomes [16]. Besides, it was found in a recent study that serum Hcy level is significantly increased in patients with GDM and closely related to decreased insulin sensitivity [17]. Increased Hcy concentration can cause autoxidation to induce and aggravate oxidative stress response, thus damaging the function of vascular endothelial cells. This can lead to microcirculatory disturbances by causing aggregation and precipitation of lipoproteins, promoting platelet aggregation and thrombosis, and participating in the pathophysiological process of decreased insulin secretion and insulin resistance in diabetics [18]. The findings in this study discovered that serum CRP, Hcy, and HOMA-IR levels in GDM patients were significantly higher than those in the control group ($p < 0.05$). There were positive correlations of serum CRP and Hcy with HOMA-IR ($p < 0.05$), indicating that inflammatory response and the increase of Hcy level may be important causes of IR, which plays an important role in the pathogenesis and progression of GDM.

AHSG is a natural inhibitor of the insulin receptor, which enhances the body's IR through suppressing the insulin receptor and hindering the signal pathway of insulin induction and activation [19]. In addition, AHSG also participates in the precipitation and transportation of calcium salts as well as resisting inflammatory reactions, and suppress tumor and vascular growth [20,21]. In this study, it was found that serum AHSG was significantly increased in patients with GDM, and positively correlated with HOMA-IR, TC, TG, and CRP, similar to the findings of a study conducted by Iyidir OT et al. [22], indicating that the inflammatory response and elevated AHSG level may cause IR and have an important role in the pathogenesis and progression of GDM.

The results of this study showed that the level of serum CRP in the GDM group was significantly increased and positively correlated with HOMA-IR, indicating that CRP may be one of the high-risk pathogenic factors of GDM and can be used as an indicator for early diagnosis of GDM. It is basically similar to the findings of a

Table 1. Comparison of basic data of patients between two groups ($\bar{x} \pm s$).

Item	GDM group (n = 72)	Control group (n = 72)	χ^2	p
Age (years old)	31.03 ± 5.24	30.92 ± 5.36	0.541	0.182
Gestational age (W)	26.17 ± 1.25	26.53 ± 1.18	0.623	0.164
Gravidity	2.1 ± 1.3	2.3 ± 0.9	2.306	0.059
Family history of DM			12.047	0.000
Yes	33	13		
No	39	59		
Family history of cardiovascular diseases			1.716	0.062
Yes	14	10		
No	58	62		

Table 2. Comparison of somatological parameters between two groups of patients ($\bar{x} \pm s$).

Item	GDM group (n = 72)	Control group (n = 72)	χ^2	p
Body weight before pregnancy (kg)	56.76 ± 7.25	55.69 ± 6.12	1.835	0.063
Height (cm)	160.78 ± 4.25	161.31 ± 4.07	0.854	0.125
BMI before pregnancy (kg/m^2)	24.03 ± 2.14	22.83 ± 1.65	2.263	0.057
Waist circumference (cm)	78.21 ± 9.36	75.49 ± 8.70	2.742	0.008
Hip circumference (cm)	94.37 ± 6.45	93.86 ± 5.74	0.715	0.256
WHR	0.84 ± 0.12	0.80 ± 0.09	3.954	0.004
Body fat (%)	32.16 ± 6.24	30.09 ± 5.46	2.018	0.013

Table 3. Comparison of glucose and lipid metabolism indexes between two groups of patients ($\bar{x} \pm s$).

Item	GDM group (n = 72)	Control group (n = 72)	χ^2	p
FPG (mmol/L)	5.42 ± 0.75	4.43 ± 0.26	8.217	0.000
1hPG (mmol/L)	9.06 ± 1.20	8.03 ± 1.07	6.624	0.000
2hPG (mmol/L)	7.95 ± 1.43	7.86 ± 1.03	0.983	0.041
HbA1c (%)	5.92±0.81	4.15 ± 0.58	3.410	0.004
FINS (mU/L)	17.38 ± 0.64	12.01 ± 1.14	8.618	0.000
HOMA-IR	4.25 ± 0.43	2.36 ± 0.29	6.751	0.000
TC (mmol/L)	6.24 ± 1.08	4.37 ± 0.85	4.412	0.002
TG (mmol/L)	3.49 ± 1.27	2.38 ± 0.64	3.125	0.006
LDL-C (mmol/L)	4.83 ± 0.91	3.45 ± 0.53	3.951	0.003
HDL-C (mmol/L)	1.12 ± 0.05	1.73 ± 0.12	2.843	0.008

study conducted by Thériault S et al. [23]. A prospective study by Qiu C et al. [24] discovered that relative risk of subsequent development of GDM in patients with an elevated serum CRP level is 3.5 times higher than that in the control group. In this study, it was uncovered that serum CRP level in the GDM group was positively correlated with HOMA-IR and TG, indicating that CRP is related to glucose and lipid metabolism disorders in patients. The mechanism may be that inflammatory factors affect or hinder insulin signal trans-

Table 4. Comparison of serum Hcy, AHSG, CRP, and 25-OH-VD of patients between the two groups ($\overline{x} \pm s$).

Item	GDM group (n = 72)	Control group (n = 72)	χ^2	p
Hcy (μmol/L)	18.12 ± 5.90	9.54 ± 3.57	4.742	0.015
AHSG (μg/L)	250.56 ± 23.12	163.84 ± 17.39	1.954	0.031
CRP (mg/L)	9.03 ± 3.17	2.41 ± 0.62	6.254	0.006
25-OH-VD (ng/mL)	15.37 ± 4.45	19.38 ± 4.72	2.715	0.026

Table 5. Correlation analysis of HOMA-IR with various serum indicators.

Biochemical indicator	r	p
FPG (mmol/L)	0.591	0.008
FINS (mU/L)	0.825	0.003
TC (mmol/L)	0.312	0.023
TG (mmol/L)	0.234	0.026
Hcy (μmol/L)	0.458	0.018
AHSG (μg/L)	0.647	0.011
CRP (mg/L)	0.487	0.016
25-OH-VD (ng/mL)	-0.323	0.021

Table 6. Correlation analysis of CRP with various serum indexes.

Biochemical indicator	r	p
HOMA-IR	0.485	0.014
TC (mmol/L)	0.331	0.017
TG (mmol/L)	0.019	0.213
Hcy (μmol/L)	0.058	0.116
AHSG (μg/L)	0.226	0.030
25-OH-VD (ng/mL)	-0.443	0.015

Table 7. Correlation analysis of AHSG with various serum indicators.

Biochemical indicator	r	p
HOMA-IR	0.647	0.011
TC (mmol/L)	0.321	0.025
TG (mmol/L)	0.314	0.028
Hcy (μmol/L)	0.058	0.116
CRP (mg/L)	0.226	0.030
25-OH-VD (ng/mL)	-0.034	0.069

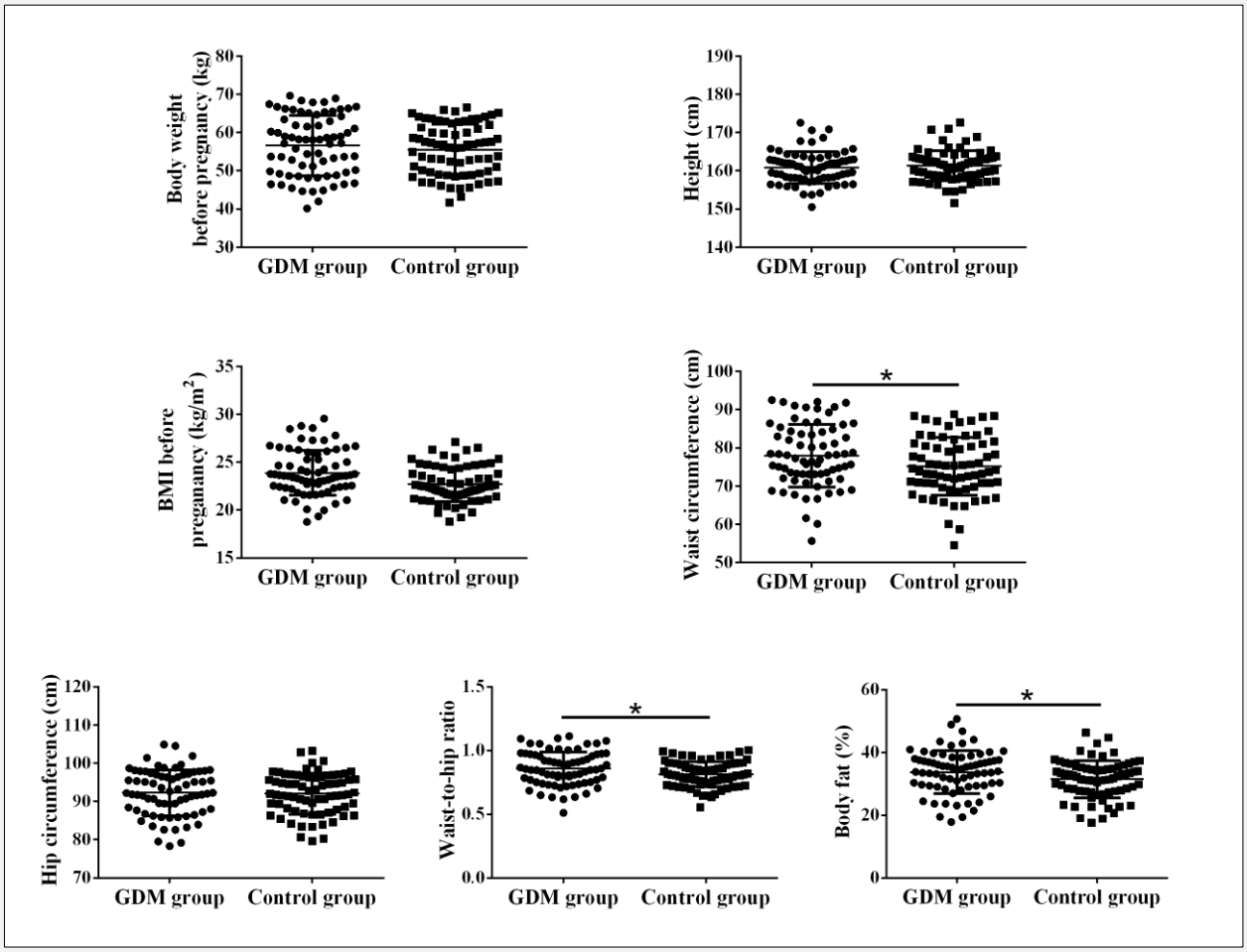

Figure 1. Comparison of somatological parameters between two groups of patients ($\bar{x}\pm s$).

*** $p < 0.05$ a statistical difference vs. control group.**

duction by activating serine/threonine kinases to form IR. IR results in increased synthesis of extremely low-density lipoprotein and reduced lipoprotein lipase activity, leading to increased concentrations of TG and LDL-C and disorders of blood lipid metabolism [25,26].

Research denoted that the low level of 25-OH-VD is associated with T2DM. Patients with T2DM have reduced blood glucose and HbA1c levels, and increased glucose tolerance, insulin secretion, and insulin sensitivity after vitamin D supplementation [27].

Serum 25-OH-VD has a longer half-life period than 1,25-hydroxyvitamin D3, which can better and more accurately reflect vitamin levels in the body. Therefore, in this study, serum 25-OH-VD was determined. The results indicated that serum 25-OH-VD level was significantly reduced in pregnant women with GDM and negatively correlated with HOMA-IR, consistent with the findings of a study by Zhang et al. [28], suggesting that the occurrence of GDM may be related to the shortage and deficiency of vitamin D, and that 25-OH-VD with a low level may have some predictive value for GDM. As a factor of high-risk pregnancy, GDM severely affects the health of the fetuses, neonates, and gravidas. The serum levels of Hcy, AHSG, and CRP in GDM patients were increased, and the level of serum 25-OH-VD was decreased, indicating that serum Hcy, AHSG, CRP, and 25-OH-VD are related to glucose and lipid metabolism disorders in GDM patients. Monitoring serum Hcy, AHSG, CRP, and 25-OH-VD may have some predictive value for GDM.

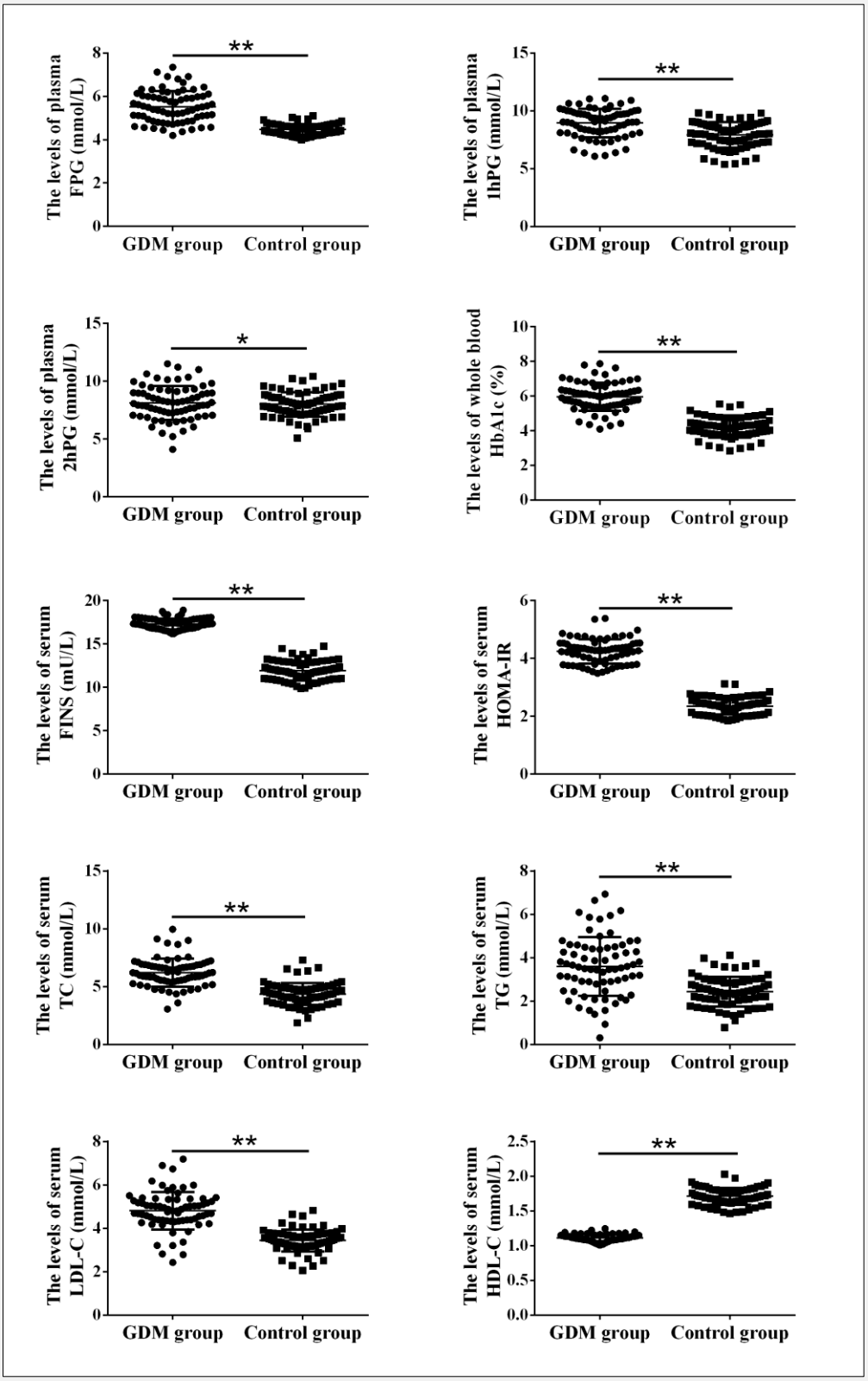

Figure 2. Comparison of glucose and lipid metabolism indexes between two groups of patients ($\bar{x} \pm s$, %).

*** $p < 0.05$ a statistically difference vs. control group, ** $p < 0.01$ a statistically difference vs. control group.**

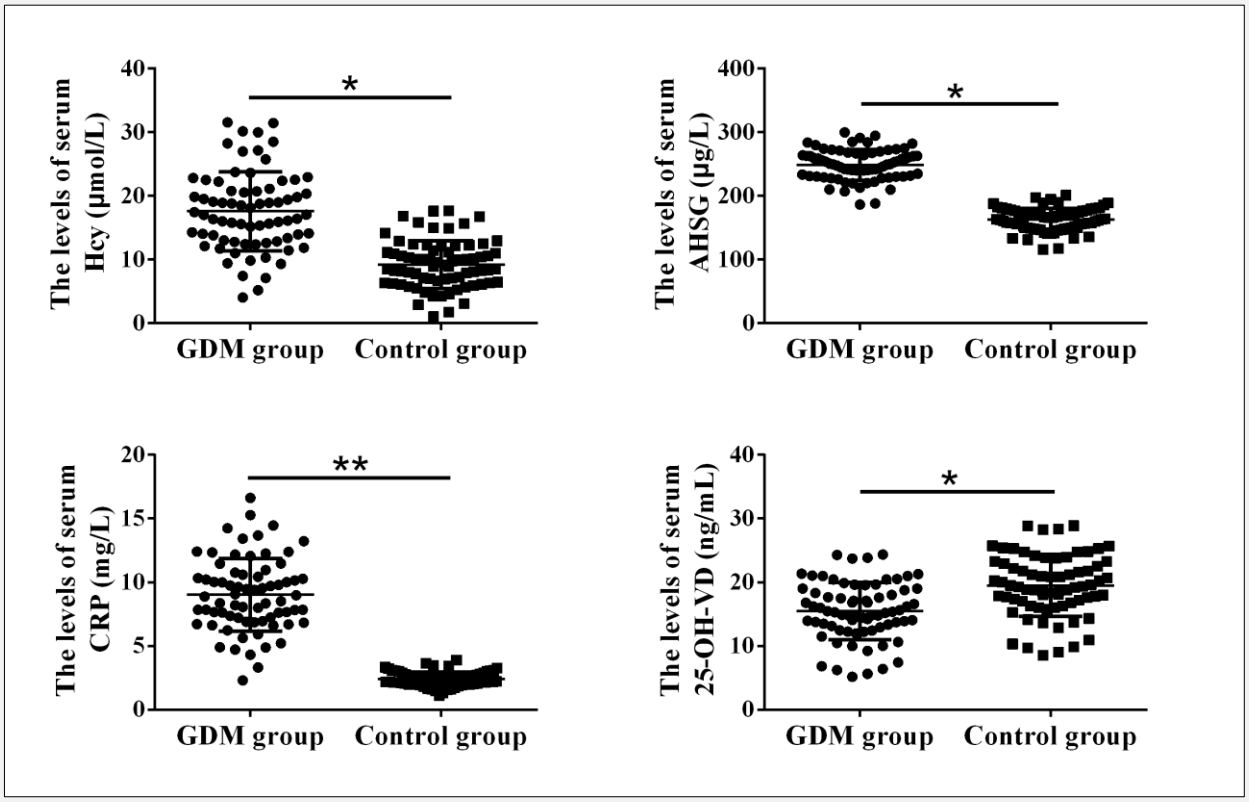

Figure 3. Comparison of serum Hcy, AHSG, CRP and 25-OH-VD of patients between the two groups ($\bar{x} \pm s$).

*** $p < 0.05$ a statistical difference vs. control group, ** $p < 0.01$ a statistical difference vs. control group.**

Declaration of Interest:

The authors have no conflicts of interest regarding the publication of this paper.

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