Elevated Levels of Serum $\beta$2-Glycoprotein I/Oxidized Low-Density Lipoprotein Complexes Are Associated with Cerebral Infarction in Patients with Type 2 Diabetes Mellitus

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Background: To determine whether the levels of $\beta$2-glycoprotein I ($\beta$2-GPI)/oxidized low-density lipoprotein (oxLDL) complexes are correlated with cerebral infarction in patients with type 2 diabetes mellitus (T2DM).

Material/Methods: The levels of $\beta$2-GPI/oxLDL complexes, oxLDL, routine lipid/lipoprotein parameters, oxidative stress molecules, and inflammatory factors were measured in 78 healthy controls, 82 diabetics without cerebral infarction, and 79 diabetics with cerebral infarction. Correlation, multiple linear regression, and logistic regression analyses were performed.

Results: Serum $\beta$2-GPI/oxLDL complexes and oxLDL levels were significantly elevated in cerebral infarction in patients with T2DM ($\beta$2-GPI/oxLDL: 1.09±0.16 U/mL; oxLDL: 47.83±8.17 mmol/L) compared with T2DM without cerebral infarction ($\beta$2-GPI/oxLDL: 0.95±0.13 U/mL; oxLDL: 41.24±7.12 mmol/L) and healthy controls ($\beta$2-GPI/oxLDL: 0.81±0.12 U/mL; oxLDL: 27.97±4.57 mmol/L). The levels of $\beta$2-GPI/oxLDL complex in lacunar infarction (1.16±0.15 U/ml) were significantly higher than atherothrombotic infarction (1.07±0.19 U/ml) and cardioembolic infarction (1.00±0.23 U/ml). In all patients with T2DM, the $\beta$2-GPI/oxLDL levels were positively correlated with total cholesterol ($r=0.474$, $p=0.001$) and triglycerides ($r=0.431$, $p=0.003$). oxLDL levels were positively correlated with total cholesterol ($r=0.445$, $p=0.002$). The logistic regression analysis indicated that elevated $\beta$2-GPI/oxLDL and oxLDL levels were independently associated with diabetic cerebral infarction.

Conclusions: Elevated levels of serum $\beta$2-GPI/oxLDL complexes are associated with cerebral infarction in patients with T2DM, especially in those with lacunar infarction.

MeSH Keywords: $\beta$2-Glycoprotein I • Cerebral Infarction • Oxidized Low-Density Lipoprotein • Type 2 Diabetes Mellitus

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Background

Diabetes mellitus is a metabolic disorder characterized by consistently elevated blood glucose and is global public health concern [1,2]. According to 2014 epidemiological data, about 8.3% of the world adult population (about 382 million people) suffers from primary type 2 diabetes (T2DM) [3]. T2DM is associated with microvascular and macrovascular complications [4] that lead to, among others problem, cardiovascular and cerebrovascular issues [5]. Particularly, cerebral infarction in people with T2DM shows a different clinical pattern compared with cerebral infarction in patients without T2DM [6]. It is estimated that the risk of cerebral infarction is increased 2–6 times in patients with T2DM compared with non-T2DM patients [7]. In addition, T2DM doubles the risk of infarction recurrence of cerebral vascular [7]. Nevertheless, intensive glycemic control has no significant impact on the occurrence of infarction, indicating that the development of cerebral infarction cannot be completely explained by the impaired glucose metabolism observed in T2DM [8].

Accumulation of multiple risk factors such as hypertension, dyslipidemia, hypercoagulability, hyperinsulinemia, accelerated fibrinolysis, and hyperglycemia lead to cerebrovascular and cardiovascular complications in diabetic patients [7,9]. It is generally recognized that T2DM-associated vascular disease is mainly associated with vascular endothelial injury and endothelial dysfunction, which are the initial steps leading to atherosclerosis. Recently, some studies analyzed the key factors of the occurrence and development of oxidative stress and inflammatory injuries in T2DM with cerebral infarction, and these factors include oxidative stress molecules and inflammatory factors [10–12] such as oxidized low-density lipoproteins (oxLDL), superoxide dismutase (SOD), malondialdehyde (MDA), glutathione peroxidase (GSH-Px), C-reactive protein (CRP), tumor necrosis factor-α (TNF-α), and interleukin (IL)-6 [13,14].

OxLDL are able to cause endothelial inflammation and endothelial dysfunction [15,16]. T2DM and cerebral infarction are both independently associated with high levels of oxidized low-density lipoproteins (oxLDL) [17–20]. Nevertheless, the exact mechanisms for the association between oxLDL and cerebral infarction are incompletely understood and oxLDL alone does not explain the entire cerebral infarction risk.

β2-glycoprotein I (β2-GPI) is a 50-kDa plasma protein that binds to negatively charged molecules, including phospholipids and heparin, and to plasma membranes of activated platelets and apoptotic cells on which phosphatidylserine is exposed [21]. oxLDL can bind to β2-GPI and form β2-GPI/oxLDL complexes [21–23], prompting macrophages to phagocytize oxLDL through the β2-GPI autoantibody-mediated pathway, thus inducing the formation of foam cells that participate in atherosclerosis and vascular diseases. It has been shown that the plasma levels of β2-GPI/oxLDL complexes are significantly increased in patients with microvascular diseases such as chronic inflammatory response and some autoimmune diseases [24–27], as well as with macrovascular diseases [28–30] and T2DM [24,31]. Nevertheless, the exact contribution of β2-GPI/oxLDL to the occurrence of cerebral infarction in T2DM is still poorly understood.

It could be hypothesized that the β2-GPI/oxLDL complexes participate in the development of cerebral infarction in patients with T2DM. Therefore, the present study aimed to determine whether the levels of β2-GPI/oxLDL complexes correlate with cerebral infarction in patients with T2DM. This could help identifying patients with T2DM at high risk of cerebral infarction.

Material and Methods

Subjects

A total of 161 newly admitted T2DM subjects were enrolled from the Department of Endocrinology of the Second Affiliated Hospital of Harbin Medical University from August 2013 to November 2014. The patients were recruited during a routine examination for their diabetes or when they consulted for symptoms of cerebral infarction. The inclusion criteria were: 1) diagnosis of T2DM according to the 1998 World Health Organization criteria [32]; 2) 18–87 years of age; and 3) underwent color duplex ultrasound of the carotids and lower extremities. The exclusion criteria were: 1) gestational diabetes mellitus; 2) diabetic macrovascular (including hypertension, cardiovascular accidents, and lower extremity vascular diseases) and microvascular complications (including diabetes nephropathy, diabetic retinopathy, and diabetic neuropathy); 3) type 1 diabetes mellitus; 4) infectious diseases; 5) chronic hepatic diseases; 6) chronic renal diseases; or 7) any genetic disease affecting lipid metabolism.

Cerebral infarction was diagnosed according to the 1989 World Health Organization criteria and to the presence of lesion ≥5 mm on magnetic resonance imaging (MRI) [33]. For the purpose of this study, the group of 79 patients with cerebral infarction was selected. Subtypes of stroke were classified according to the Trial of Org 10172 in Acute Stroke Treatment (TOAST) [34]. Subtypes of cerebral infarction included atherothrombotic infarction (n=26), lacunar infarction (n=33), cardioembolic infarction (n=16), infarction of undetermined origin (n=1), and infarction of unusual cause (n=3). Lacunar infarction accounts for more than 25% of brain infarction [35]. The undetermined origin and unusual cause are regarded as other subtypes due to the small sample size.
The patients with T2DM were divided into those with cerebral infarction and those without any clinical signs and symptoms of diabetic cerebral infarction. In addition, 78 healthy outpatients who visited the Second Hospital affiliated to Harbin Medical University for routine health examination were selected as the control group and were age- and sex-matched. They all had normal physical, electrocardiography, ultrasound examination, and biochemical examinations. The exclusion criteria were: 1) hyperlipidemia; 2) hypertension; 3) cardiovascular or cerebrovascular diseases; 4) diabetes mellitus; 5) severely impaired hepatic function; or 6) any recent surgery.

The study protocol was approved by the Ethics Committee of Harbin Medical University. All subjects provided a written informed consent.

**Blood sampling**

Blood samples were collected into a routine tube after a 10-h overnight fast centrifuged immediately at 4°C for 10 min at 3000 g, and stored at −80°C.

**β2-GPI/oxLDL measurement**

β2-GPI/oxLDL was measured using a sandwich enzyme-linked immunosorbent assay, as previously described and without any modification [31]. The polyclonal antibody against human β2-GPI was coated onto 96-microwell plates and the polyclonal anti-human apoprotein B antibody was used as the detection antibody [36]. Serum sample (500 μL) containing 10 μmol/L MgCl₂, were incubated for 2 h at room temperature. Then, polyethylene glycol (PEG) was added and the samples were incubated at 4°C overnight. The samples were centrifuged at 10,000 rpm for 20 min. The precipitates were resuspended with 500 μL washing solution containing 0.5% gelatin and 0.05% Tween-20 in 0.01 mol/L PBS buffer solution. Antihuman β2-GPI antibody was added to microwells at 2.5 μg/ml (100 μL/well) and incubated for 2 h at 37°C and then overnight at 4°C. Samples diluted at 1: 40 (resuspended with gelatin in PBST) or serial reference sera were added to the wells and incubated for 2 h after blocking with 1% gelatin in 0.01 mol/L PBS. Then, the wells were incubated with BSA for 2 h. Standard β2-GPI/ox-LDL complexes were added in the wells and incubated at 4°C overnight in order to obtain the standard curve. HRP-labeled goat anti-rabbit LDL polyclonal antibody was added to the wells and incubated at room temperature for 3 h. Color was developed with TMBUS substrate and the reaction was stopped by 2 M sulfuric acid. The absorbance was read at 450 nm with a microplate reader. Between each step, the wells were washed 3 times with PBS containing 0.05% Tween 20. The reference serum β2-GPI/oxLDL was made using a pooled fresh plasma sample from 60 healthy subjects. Serum β2-GPI/oxLDL complexes concentration (expressed in U/mL) was calculated against a reference curve built with serial dilutions of the reference serum. Intra- and inter-assay coefficients of variation were all <15%.

**Biochemistry**

CRP, TNF-α, and IL-6 were measured by radioimmunoassay (Belfang Technology, Beijing, China). SOD, MDA, and GSH-Px were measured with commercial kits using enzymatic methods (Jiancheng Technology, Nanjing, China). Fasting plasma glucose (FPG), 2-h postprandial blood glucose (2hPBG), total cholesterol (TC), HDL-cholesterol, LDL-cholesterol, and triglycerides (TG) were determined using a ROCHE Modular P800 Automatic Biochemical Analyzer (Roche Diagnostics, Basel, Switzerland). Serum insulin levels were measured using a Siemens centaur XP Chemiluminescence Immune Analyzer (Siemens, Erlangen, Germany). HbA1c was measured using a Varint II Turbo (Bio-Rad, Hercules, CA, USA).

**Statistical analysis**

Continuous data were tested for normal distribution using the Kolmogorov-Smirnov test. Normally distributed continuous data are presented as mean ± standard deviation and analyzed by ANOVA with the LSD post hoc test. Non-normally distributed variables are presented as median (range) and analyzed using the Kruskal-Wallis test with the post hoc Mann-Whitney U test. The Spearman correlation analysis was used to determine the correlations between parameters. Univariate and multivariate logistic regression analyses were used to analyze the relative risks for selected variables. All statistical analyses were performed with SPSS 16.0 (IBM, Armonk, NY, USA). Two-sided P-values <0.05 were considered statistically significant.

**Results**

**Characteristics of the subjects**

Table 1 presents the characteristics of the subjects. There were no differences in age or sex among the 3 groups (all P>0.05). Compared with controls, diabetics had higher TC, TG, low-density lipoprotein cholesterol (LDL-C), FBG 2hPG, HbA1c, FINS, and CRP, and lower high-density lipoprotein cholesterol (HDL-C) (all P<0.05). Compared with diabetics without cerebral infarction, diabetics with cerebral infarction had higher TC and TG, and lower 2hPG (all P<0.05).

**Serum β2-GPI/oxLDL and oxLDL**

Compared with the control group (β2-GPI/oxLDL complexes: 0.81±0.12 U/mL; oxLDL: 27.97±4.57 mmol/L), serum β2-GPI/oxLDL and oxLDL levels were higher in patients with T2DM;
furthermore, serum β2-GPI/oxLDL and oxLDL levels were higher in cerebral infarction in patients with T2DM (β2-GPI/oxLDL complexes: 1.09±0.16 U/mL; oxLDL: 47.83±8.17 mmol/L) compared with patients with T2DM but without cerebral infarction (β2-GPI/oxLDL complexes: 0.95±0.13 U/mL; oxLDL: 41.24±7.12 mmol/L) (Table 1).

Table 1. Biochemical characteristics of the patients with T2DM and controls.

| Variables          | Control (n=78) | Diabetics without cerebral infarction (n=82) | Diabetics with cerebral infarction (n=79) |
|--------------------|----------------|----------------------------------------------|-----------------------------------------|
| β2-GPI/oxLDL (U/ml)| 0.81±0.12      | 0.95±0.13‡‡‡‡                                   | 1.09±0.16‡‡‡‡                               |
| oxLDL (mmol/L)     | 27.97±4.57     | 41.24±7.12‡‡‡‡                                 | 47.83±8.17‡‡‡‡                             |
| Age (years)        | 56.1±6.9       | 57.2±6.1                                     | 58.9±8.5                                  |
| Male, n (%)        | 40 (51.3%)     | 42 (53.2%)                                   | 41 (51.3%)                                |
| TC (mmol/L)        | 4.37±0.43      | 4.73±0.96                                    | 5.32±1.13‡‡‡‡                             |
| TG (mmol/L)        | 1.10 (0.85,1.34)| 1.56 (1.33,2.76)§§‡‡‡‡                     | 2.63 (1.93,5.03)‡§‡‡‡                  |
| HDL-C (mmol/L)     | 1.37±0.18      | 1.11±0.23‡‡‡‡                                 | 1.21±0.23‡†                                |
| LDL-C (mmol/L)     | 2.43±0.40      | 2.95±0.79‡§‡‡‡                                 | 3.16±0.83‡§‡‡‡                             |
| FBG (mmol/L)       | 5.15 (4.86,5.31)| 7.66 (6.69,8.99)§§‡‡‡‡                     | 7.17 (6.49,8.93)§§‡‡‡‡                  |
| 2hPG (mmol/L)      | 6.62 (6.31,7.07)| 17.47 (13.74,20.24)§§‡‡‡‡                 | 15.42 (12.43,17.18)§§‡‡‡‡               |
| HbA1c (%)          | 5.3 (5.1,5.6)  | 7.55 (6.28,9.13)§§‡‡‡‡                     | 8.65 (6.3,7.56)§§‡‡‡‡                 |
| FINS (μU/ml)       | 6.50 (5.33,6.85)| 6.85 (4.98,13.65)§                      | 7.45 (5.65,13.2)‡§‡‡‡                  |
| IL-6 (pg/ml)       | 129.91±70.27   | 127.34±60.79                                 | 129.71±95.95                             |
| TNF-α (pg/ml)      | 1.36 (0.76,2.22)| 1.83 (1.29,2.73)§§                          | 1.68 (1.08,2.62)‡†                     |
| CRP (μg/ml)        | 0.93 (0.76,1.21)| 1.49 (1.0,2.44)‡                      | 1.37 (1.1,1.71)‡§‡                            |
| SOD (U/ml)         | 116.63±38.11   | 97.14±45.60                                  | 94.70±83.90                              |
| GSH (nmol/ml)      | 351.14±278.24  | 357.49±183.36                                | 383.10±208.72                            |
| MDA (nmol/ml)      | 36.64±40.29    | 43.25±54.52                                  | 44.22±65.63                              |

§ Compared with controls, P<0.05; ‡‡‡‡ Compared with controls, P<0.01; ‡§‡§ Compared with group 1, P<0.05; **‡‡‡‡ Compared with group 1, P<0.01. β2-GPI = β2-glycoprotein I; oxLDL = oxidized low-density lipoprotein; TC = total cholesterol; TG = triglycerides; LDL-C = low-density lipoprotein cholesterol; HDL-C = high-density lipoprotein cholesterol; FBG = fasting blood glucose; 2hPG = 2-h postprandial glucose; FINS = fasting insulin; IL = interleukin; TNF-α = tumor necrosis factor α; CRP = C-reactive protein; SOD = superoxide dismutase; GSH = glutathione peroxidase; MDA = malondialdehyde.

Associations among β2-GPI/oxLDL, oxLDL, lipids, oxidative stress molecules, and inflammatory factors in patients with T2DM

Among all T2DM patients, the β2-GPI/oxLDL complexes levels were positively correlated with TC (r=0.474, P=0.001) and TG (r=0.431, P=0.003). oxLDL levels were positively correlated with TC (r=0.445, P=0.002) (Table 2).
### Table 2. Correlations among β2-GPI/oxLDL, oxLDL, and other parameters in T2DM patients.

| Variables group | Statistics | β2-GPI/oxLDL | oxLDL |
|-----------------|------------|--------------|-------|
| TC              | r Value    | 0.474**      | 0.445** |
|                 | p Value    | 0.001        | 0.002 |
| TG              | r Value    | 0.431**      | 0.206 |
|                 | p Value    | 0.003        | 0.170 |
| HDL-C           | r Value    | 0.179        | 0.074 |
|                 | p Value    | 0.234        | 0.627 |
| LDL-C           | r Value    | 0.098        | -0.005 |
|                 | p Value    | 0.516        | 0.976 |
| FBG             | r Value    | -0.070       | -0.045 |
|                 | p Value    | 0.645        | 0.099 |
| 2hPG            | r Value    | -0.044       | -0.068 |
|                 | p Value    | 0.771        | 0.652 |
| HbA1c           | r Value    | 0.101        | 0.021 |
|                 | p Value    | 0.505        | 0.889 |
| FINS            | r Value    | 0.010        | 0.071 |
|                 | p Value    | 0.946        | 0.641 |
| IL-6            | r Value    | -0.130       | -0.250 |
|                 | p Value    | 0.390        | 0.093 |
| TNF-α           | r Value    | 0.130        | 0.155 |
|                 | p Value    | 0.390        | 0.302 |
| CRP             | r Value    | -0.044       | -0.062 |
|                 | p Value    | 0.773        | 0.681 |
| SOD             | r Value    | -0.093       | -0.168 |
|                 | p Value    | 0.540        | 0.266 |
| GSH             | r Value    | -0.042       | -0.103 |
|                 | p Value    | 0.782        | 0.497 |
| MDA             | r Value    | -0.203       | -0.111 |
|                 | p Value    | 0.177        | 0.463 |
| β2-GPI/oxLDL    | r Value    | 1.000        | 0.563** |
|                 | p Value    | <0.001       |      |
| oxLDL           | r Value    | 0.563**      | 1.000 |
|                 | p Value    | <0.001       |      |

β2-GPI – β2-glycoprotein I; oxLDL – oxidized low-density lipoprotein; TC – total cholesterol; TG – triglycerides; LDL-C – low-density lipoprotein cholesterol; HDL-C – high-density lipoprotein cholesterol; FBG – fasting blood glucose; 2hPG – 2-h postprandial glucose; FINS – fasting insulin; IL – interleukin; TNF-α – tumor necrosis factor α; CRP – C-reactive protein; SOD – superoxide dismutase; GSH – glutathione peroxidase; MDA – malondialdehyde.
The univariate analyses revealed that increased \( \beta_2 \)-GPI/oxLDL and oxLDL levels were significantly associated with the presence of T2DM with (\( \beta_2 \)-GPI/oxLDL, OR=2.48, 95% CI: 1.72–3.03, \( P=0.001 \); oxLDL, OR=2.17, 95% CI: 0.98–4.8, \( P=0.045 \)) or without (\( \beta_2 \)-GPI/oxLDL, OR=1.61, 95% CI: 1.11–3.77, \( P=0.004 \); oxLDL, OR=1.46, 95% CI: 1.17–1.83, \( P=0.001 \)) cerebral infarction (Table 3).

In the multivariate analysis, after adjusting for age, gender, and serum lipid levels, \( \beta_2 \)-GPI/oxLDL (OR=3.11, 95% CI: 2.01–5.32, \( P=0.033 \)) and oxLDL (OR=2.91, 95% CI: 1.41–4.45, \( P=0.022 \)) were independently associated with the presence of cerebral infarction. Only high oxLDL levels were independently associated with T2DM without complications (OR=1.26, 95% CI: 0.96–2.14, \( P=0.042 \)), respectively.

**Associations of \( \beta_2 \)-GPI/oxLDL and oxLDL with cerebral infarction in patients with T2DM**

The levels of \( \beta_2 \)-GPI/oxLDL complex in lacunar infarction (1.16±0.15 U/ml) were significantly higher than atherothrombotic infarction (1.07±0.19 U/ml) and cardioembolic infarction (1.00±0.23 U/ml). It was considered statistically significant \((P<0.05)\). Compared with other subtypes (0.99±0.06 U/ml), the levels of the complex were higher in the lacunar infarction group. There was no statistically significant \((P>0.05)\) (Table 4).

**Discussion**

The present study showed that serum \( \beta_2 \)-GPI/oxLDL complexes and oxLDL levels were significantly elevated in cerebral infarction in patients with T2DM, compared with T2DM without cerebral infarction and healthy controls. The \( \beta_2 \)-GPI/oxLDL levels were independently correlated with oxLDL in T2DM patients. Elevated \( \beta_2 \)-GPI/oxLDL complexes and oxLDL levels were independently associated with diabetic cerebral infarction.
Hyperglycemia can cause decreased erythrocyte deformability, enhanced blood platelet viscosity, increased thromboxane B2, decreased prostacyclin levels, and thickened vessel wall, leading to microcirculation disorder and higher risk of cerebral infarction [37]. In addition, hyperglycemia increases mortality and leads to poor functional recovery in both diabetic and non-diabetic patients after stroke [38]. Moreover, the recurrence rate and morbidity of T2DM complicated with acute cerebral infarction are high, with poor prognosis [39]. Therefore, predicting the presence of diabetic cerebral infarction is very important.

OxLDL are recognized by scavenger receptors on macrophages, leading to the formation of foam cells that constitute the core of atherosclerotic plaques [36,40,41]. β2-GPI binds to lipoproteins and participates in lipid metabolism, and has anticoagulant activity. Blood oxLDL can be decomposed by hepatic interstitial cells [42], but the negatively charged oxLDL may interact with β2-GPI and form neutrally charged β2-GPI/oxLDL complexes [43], which can avoid scavenger cell recognition and stably exist. Recently, studies have shown that β2-GPI/oxLDL levels were significantly increased in patients with autoimmune diseases such as rheumatoid arthritis and systemic lupus erythematosus or chronic diseases including coronary artery disease [44]. T2DM is associated with autoimmune features and inflammatory state [1,2,5]. In this study, we found that β2-GPI/oxLDL levels were increased in cerebral infarction in patients with T2DM. The β2-GPI/oxLDL and oxLDL levels in T2DM patients were independent from each other. Elevated β2-GPI/oxLDL and oxLDL levels were independently associated with cerebral infarction in T2DM. High oxidative stress occurs at the early stage of diabetes [45], leading to high levels of oxLDL [46,47]. Compared with β2-GPI/oxLDL levels, oxLDL levels may have a more obvious association with the presence of T2DM without complications.

Inflammatory cytokines play an important role in the occurrence and development of diabetes and atherosclerosis. CRP is an acute phase protein and represents the systemic inflammatory status [48]. CRP stimulates the vascular smooth muscle cell and is detrimental to the endothelial function [49]. The causes of increased levels of inflammatory factors such as serum CRP in diabetic patients are associated with insulin resistance and long-term chronic inflammatory stimulation by hyperglycemia [50]. Consistently, the present study showed that patients with T2DM had higher CRP levels than controls, but there was no difference between the 2 T2DM groups.

It is well established that oxidative stress is involved in the pathogenesis of diabetic complications. Oxidative stress is due to imbalance between production of reactive oxygen species (ROS) and antioxidant defense [51]. β2-GPI/oxLDL are the product of this imbalance. Nevertheless, the present study did not show any difference in SOD, GSH, or MDA, which all play roles in oxidative stress and associated diseases. Additional studies are necessary to determine whether elevated β2-GPI/oxLDL levels result from long-term accumulation of stable complexes, or whether other factors are involved.

It is well known that diabetes accelerates the clinical course of atherosclerosis and contributes to cerebrovascular recurrence and increased cardiovascular morbidity and mortality [6]. The prevalence of diabetes mellitus is higher in lacunar infarction than in the other subtypes of stroke, and it is a significant factor for lacunar infarction, particularly in the cases of multiple lacunar infarction [52]. In our ischemic stroke subtype groups, elevated β2-GPI/oxLDL levels might be associated with presence in T2DM patients with different subtypes of cerebral infarction. The β2-GPI/oxLDL levels were increased in lacunar and in non-lacunar ischemic stroke, especially in those with lacunar infarction. We found higher occurrence of lacunar infarction compared with atherothrombotic infarction and cardioembolic infarction. In the next clinical study, we should focus on the role of different risk profiles in stroke subtypes.

This study has some limitations, including the small cohort size and the recruitment of patients from a single center. In addition, cerebral infarction was diagnosed based on MRI findings in symptomatic patients, and it is possible that cerebral infarction was present in patients without symptoms. Finally, the study was not designed to address any causality issue. These results should be confirmed with multicenter studies and with a larger number of subjects. The findings should be interpreted with caution and considered as hypotheses-generating.

**Conclusions**

This study showed that serum β2-GPI/oxLDL and oxLDL levels were increased in patients with T2DM, especially in those with cerebral infarction. β2-GPI/oxLDL and oxLDL levels in T2DM patients were independently correlated. Elevated β2-GPI/oxLDL levels might be associated with the presence of diabetic cerebral infarction complications. These findings may provide important clues for the understanding of the pathogenic role of circulating β2-GPI/oxLDL in T2DM and the subtypes of cerebral infarction.

**Conflict of interests**

None.
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