A comparative study on C-reactive protein and gamma-glutamyl transferase as novel inflammatory markers of type 2 diabetes mellitus

Tushar K. BANDYOPADHYAY[1]

[1] Purulia Govt. Medical College, Purulia, West Bengal, India

ABSTRACT

Studies in the last few years have linked oxidative stress and inflammation to beta-cell function resulting from chronic exposure to hyperglycemia. Recent prospective trials have suggested that an elevated level of C-reactive protein and gamma-glutamyl transferase enzyme is associated with subsequent development of diabetes.

The present study aimed to examine the relationship between gamma-glutamyl transferase and the marker of inflammation C-reactive protein in patients with diabetes.

The study was conducted on 300 patients, including 100 healthy controls and 200 patients with type 2 diabetes. Plasma glucose levels (fasting and postprandial), serum levels of high-sensitivity C-reactive protein, levels of glycosylated hemoglobin, and serum gamma-glutamyl transferase hepatic enzyme levels were measured.

The mean high-sensitivity C-reactive protein and gamma-glutamyl transferase levels in patients with type 2 diabetes mellitus were significantly higher than the values in controls (P < 0.0010). Further, a significant positive correlation was observed between gamma-glutamyl transferase levels and high-sensitivity C-reactive protein levels in patients with type 2 diabetics (r = 0.312, P = 0.001).

The increase in the levels of high-sensitivity C-reactive protein and gamma-glutamyl transferase in patients with diabetes and their significant association might be a result of inflammation and oxidative stress in diabetes mellitus.

Key words: C-reactive protein; Gamma-glutamyl transferase; inflammation; oxidative stress, type 2 diabetes

INTRODUCTION

Research in the last few years has linked oxidative stress (OS) and inflammation to β-cell dysfunction resulting from chronic exposure to hyperglycemia (1, 2), free fatty acids, or a combination of the two. A growing body of data (3) reinforces the concept that inflammation also plays an important role in the pathogenesis of type 2 diabetes mellitu (DM) and links DM with concomitant conditions with inflammatory components.

DM is a group of metabolic diseases characterized by hyperglycemia resulting from defects of insulin secretion, insulin action, or both. Type 2 DM is caused by a combination of resistance of insulin action and an inadequate compensatory insulin secretory response. This form of DM accounts for approximately 90%–95% of those with DM and was previously referred to as noninsulin-dependent DM (NIDDM) or adult-onset DM.

Prospective studies have described that a high level of gamma-glutamyl transferase (GGT) is associated with the subsequent development of diabetes. Recently, serum GGT has been recognized as a marker of oxidative stress. Indeed oxidative processes are key components of chronic inflammation acting on multiple pathways and amplifying inflammatory reactions. Further activation of inflammatory processes may contribute to the development of type 2 DM (5-7).

C-reactive protein (CRP) is considered to be a major inflammatory cytokine that functions as a nonspecific defense mechanism in response to tissue injury or infection. Recent prospective studies have suggested a relationship between an elevated level of CRP an increasing risk of developing type 2 DM (8-10).

Correspondence:
Tushar Kanti BANDYOPADHYAY
Purulia Govt. Medical College, Purulia, West Bengal, India.
e-mail: drtkban011@gmail.com

Since oxidative stress appears to be a key component of many reactions associated with chronic inflammation, studying the levels of GGT (a liver enzyme) and CRP (an inflammatory marker) in patients with diabetes was found to be interesting. Therefore, the aim of this study was to compare the levels of GGT and the marker of inflammation (CRP) in patients with diabetes.

MATERIALS AND METHODS

The study was conducted from 2014 to 2018 at Medical College Kolkata, Midnapore Medical College Hospital, Midnapore, and Purulia Govt. Medical College Purulia (Indoor and Outpatient department of Medicine). A total of 300 participants were enrolled for this study, including 100 healthy controls (50 male and 50 female with a mean age of 53.8 ± 8.63 years) and 200 patients with type 2 diabetes (75 male and 70 female with a mean age of 10.6 ± 2.78 years).

The local ethics committee approved the study. Before participation, the volunteers were fully informed of the nature and purpose of the study and written consent was obtained from each.

Inclusion criteria

- Type 2 DM was diagnosed on the basis of American Diabetic association 2008 criteria (fasting plasma glucose ≥ 126.0 mg/dL) after repeat testing.
- All participants were nonalcoholics and nonsmokers.
- The patients with diabetes did not receive any other medications other than oral antidiabetic drugs.
- Among the baseline parameters, systolic blood pressure and diastolic blood pressure (SBP and DBP) were measured three times and averaged.
- All participants were hypertensive.

Exclusion criteria

Participants with nutritional deficiency, estrogen therapy, or active inflammatory diseases were excluded from the study.

Anthropometric measurements

Height without shoes and weight were measured, and body mass index (BMI) was calculated as kilogram divided by the square of height in meters.

Biochemical measurements

Fasting blood samples were drawn from all participants and analyzed. Plasma glucose (fasting and postprandial) concentrations were measured using glucose oxidase–peroxidase, serum high-sensitivity CRP (hs-CRP) levels were measured using the immunoturbimedric method, glycosylated hemoglobin levels were analyzed by the cation exchange resin method, and serum GGT hepatic enzyme levels were measured using the modified kinetic colorimetric method.

Statistical evaluation

Data were expressed as mean $\pm$ standard deviation.

The means were compared using the Student t test. The Pearson's correlation analysis was used for the correlation of parameters measured. The analysis was two tailed, and a P value ≤ 0.05 was considered as statistically significant.

RESULTS

The baseline characteristics of patients with type 2 diabetes and controls are given in Table 1. The baseline clinical characteristics, such as BMI, age, and blood pressure (SBP/DBP) did not differ in patients with type 2 diabetes and controls ($P = 0.21$, $P = 0.11$, $P = 0.87/0.71$). The mean fasting plasma glucose, postprandial glucose and Glycated Hemoglobin (GHb) levels of patients with type 2 diabetes were significantly higher than those in controls ($P < 0.0001$) (Table 1).

The mean hs-CRP levels in patients with type 2 diabetes (2.42 ± 1.01 mg/dL) were higher than those in controls (1.15 ± 0.37 mg/dL) and were found to be statistically significant ($P < 0.001$) (Table 2).

A similar trend was observed in GGT values in patients with type 2 diabetes compared with controls ($P < 0.001$) (Table 2).

Further, a significant positive correlation was observed between GGT and hs-CRP in patients with type 2 DM ($r = 0.312$, $P = 0.001$) (Table 3). However, the association of GGT and hs-CRP was nonsignificant in controls ($r = 0.041$, $P = 0.78$) (Table 3).

The values for BMI, age, and SBP/DBP did not differ significantly in male and female controls and male and female patients with diabetes. However, male and female patients with type 2 diabetes had significantly higher values of fasting glucose, postprandial glucose, and glycosylated hemoglobin compared with male and female controls ($P < 0.0001$) (Table 1).

TABLE 1: Baseline characteristics of healthy controls and patients with type 2 diabetes.

	BMI (kg/m²)	Age (year)	SBP/DBP (mm hg)	FPG (mg/dL)	PPG (mg/dL)	GHb (%)
Healthy controls (n = 100)	23.1 ± 3.99	53.8 ± 8.63	128/77.2	83.4 ± 13.3	104.0 ± 9.95	6.18 ± 0.31
Patients with type 2 diabetes (n = 200)	24.0 ± 3.70	51.1 ± 10.01	129/76.0	159.4 ± 48.4	237.0 ± 69.5	10.2 ± 1.36
P value	0.21 (NS)	0.11 (NS)	0.87/0.71 (NS)	<0.0001 (HS)	<0.0001 (HS)	<0.0001 (HS)
Male	±			±		
Healthy controls (n = 50)	24.4 ± 3.7	54.4 ± 8.85	128/76.9	84.1 ± 13.4	105.0 ± 11.9	6.13 ± 0.30
Patients with type 2 diabetes (n = 100)	23.44 ± 3.5	50.2 ± 10.3	1294 ± 76.5	151.0 ± 44	232.0 ± 72.6	10.1 ± 1.39
P value	0.39	0.38	0.78/0.81	<0.00001	<0.0001	0.0001
Female						
Healthy controls (n = 50)	23.6 ± 3.65	53.1 ± 9.78	128/77.4	82.6 ± 33.5	102.0 ± 7.65	6.23 ± 0.31
Patients with type 2 diabetes (n = 100)	23.5 ± 3.7	52.1 ± 99.80	128/77.1	167.0 ± 51.6	242.0 ± 66.7	10.3 ± 1.33
P value	0.36 (NS)	0.64 (NS)	0.95/0.84 (NS)	<0.0001 (HS)	<0.0001 (HS)	<0.0001 (HS)
Female versus male						
Healthy controls P value	0.39 (NS)	0.62 (NS)	1.0/0.78 (NS)	0,62 (NS)	0.41 (NS)	0.25 (NS)
Patients with type 2 diabetes P value	0.27 (NS)	0.34 (NS)	0.68/0.64 (NS)	0.10 (NS)	0.50 (NS)	0.34 (NS)

TABLE 2: Values of high-sensitivity C-reactive protein (hs-CRP) and gamma-glutamyl transferase in participants.

Participants	Hs-CRP (mg/L)	GGT (U/L)
Healthy controls	1.15 ± 0.37	27.6 ± 7.36
Males (*n* = 50)	1.08 ± 0.34	33.0 ± 5.53
Females (*n* = 50)	1.12 ± 0.33	31.2 ± 4.40
P value	0.68 (NS)	0.21 (NS)
Patients with type 2 diabetes	2.42 ± 1.01	35.4 ± 8.90
Males (n = 100)	2.32 ± 0.99	32.8 ± 9.21
Females (n = 100)	2.45 ± 1.02	37.9 ± 7.86
P value	0.57	0.003 (S)
Healthy controls versus patients with diabetes P value	<0.0001 (HS)	<0.001 (HS)

TABLE 3: Pearson's correlation analysis of the values of high-sensitivity C-reactive protein (hs-CRP) and gamma-glutamyl transferase (GGT) in participants.

Participants	Parameters correlated	
Patients with type 2 diabetes	Hs-CRP	GGT
r value	0.312	
P value	0.001 (HS)	
Healthy controls		
r value	0.041	
P value	0.78 (NS)	

HS: Highly significant; NS: Not significant.

DISCUSSION

In the present study, a significant high ($P < 0.001$) increase in serum GGT levels was observed in patients with type 2 diabetes compared with healthy controls. The results were in accordance with the findings of many prospective studies where a strong relationship between GGT concentration and the incidence of diabetes was observed in nonalcoholics independently of classical cardiovascular risk factors (8,9,16). The results of this study also indicated a significant increase in the values of hs-CRP ($P = 0.00010$) in patients with diabetes compared with healthy controls.

Various factors are considered in the pathogenesis of diabetes. Substantial evidence indicates that OS increases in patients with diabetes and glucose can substantially contribute to the increased production of reactive oxygen species (ROS) (11). Beta cells of pancreas and vascular endothelium possibly have the lowest potential for scavenging oxygen free radicals. This suggests the susceptibility of beta cells to oxidative stress (17,18).

It has been speculated that elevated GGT levels might be a defensive response to oxidative stress or a marker of OS is involved directly in the generation of ROS, especially in the presence of iron and other transition metals inducing lipid peroxidation in human biological membranes (17,18).

Activation of inflammatory processes may contribute to the development of type 2 DM. Recent prospective studies have suggested that an elevated level of CRP is associated with an increased risk of developing type 2 DM (8-10).

A significant correlation between GGT and hs-CRP was observed in patients with diabetes in the present study ($r = 0.312$, $P = 0.0001$). Oxidative stress processes might have an implication in chronic inflammation (11). It is hypothesized that elevation in the hs-CRP level and the related oxidative stress gives rise to a subsequent inflammatory response.

In the present study, patients with type 2 diabetes having complications were not considered and hence GGT levels were not assessed in these patients. However, several population studies (19,20) have shown a cross-sectional association between serum GGT concentrations and many cardiovascular risk factors or components of insulin resistance syndrome, including age, obesity, smoking, lack of exercise, blood pressure, dyslipidemia, and DM. In addition, prospective studies have shown baseline serum GGT concentration as an independent risk factor for the development of cardiovascular or cerebrovascular diseases. Thus, it is presumed that OS may be the underlying cause of any of the complications of diabetes and it amplifies inflammatory processes too. Hence, the levels of GGT and hs-CRP may serve as biomarkers for evaluating diabetic complications in future studies.

In conclusion, the rise in the levels of hs-CRP and GGT in patients with diabetes and their significant association might be a result of oxidative stress caused by diabetes, The follow-up of these patients with diabetes may further reflect the complications of DM.

This study proposed an association between GGT and hs-CRP, which might be considered a limitation of the study. However, the study may be extended and carried out on a large cohort to establish a causative relationship between GGT and hs-CRP levels.

Only patients with diabetes having no complications were considered in this study, which is another limitation of this study.

However, the findings may be important to assess GGT and hs-CRP levels in patients with type 2 diabetes having complications and evaluate the severity of complications (both micro- and macrovascular) in the future. Thus, GGT and hs-CRP levels may be considered sensitive biomarkers in predicting diabetes, as discussed in the present study, and in evaluating the degree of complications in patients with diabetes in future studies.

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