

Original Research Article

To study levels of serum fibrinogen in type 2 diabetes mellitus and its association with diabetic microvascular complications

Gurinder Mohan*, Ranjeet Kaur, Aakash Aggarwal, Parminder Singh

Department of Medicine, Sri Guru Ram Das Institute of Medical Sciences and Research, Amritsar, Punjab, India

Received: 28 December 2016

Accepted: 08 January 2017

*Correspondence:

Dr. Gurinder Mohan,

E-mail: drgurinder1968@gmail.com

Copyright: © the author(s), publisher and licensee Medip Academy. This is an open-access article distributed under the terms of the Creative Commons Attribution Non-Commercial License, which permits unrestricted non-commercial use, distribution, and reproduction in any medium, provided the original work is properly cited.

ABSTRACT

Background: Diabetes mellitus is a hypercoagulable state associated with atherosclerosis leading to development of vascular complications, including microvascular complications.

Methods: In our study a total of 60 diabetic patients with duration of diabetes more than 5 years, attending the OPD/indoor of SGRDIMSAR, Amritsar, Punjab, India were included. They were divided in two groups, group A of 30 patients including diabetics with any of the three microvascular complications (diabetic nephropathy, diabetic retinopathy and diabetic neuropathy) and group B of 30 patients including diabetics without any microvascular complication. Group C comprised of 30 age and sex matched non-diabetic subjects who served as controls. Subjects with liver cirrhosis, malignancy or coagulation disorder were excluded. After taking the consent, detailed history taking and detailed physical examination and relevant investigations were done. The serum fibrinogen (hemostasis marker), HBA1C and UACR (urine albumin creatinine ratio) along with routine investigations were measured.

Results: It was observed that serum fibrinogen levels were significantly higher in diabetic patients (266.16 ± 54.73 mg/dl) as compared to non-diabetic controls (174.66 ± 18.32 mg/dl); $p < 0.001$. Further, serum fibrinogen levels were found to be significantly higher in diabetic patients with microvascular complications (293.43 ± 51.09 mg/dl) as compared to those without microvascular complications (238.90 ± 44.12); $p < 0.001$.

Conclusions: Significantly high serum fibrinogen level was found in diabetic patients as compared to controls and was in positive correlation with development of microvascular complications.

Keywords: Microvascular complications, Serum fibrinogen

INTRODUCTION

Diabetes mellitus is a chronic, progressive disease characterized by elevated levels of blood glucose, which occurs either when the pancreas does not produce enough insulin, or when the body cannot effectively use the insulin.¹ It is associated with reduced life expectancy, significant morbidity due to specific diabetes related microvascular complications (nephropathy, retinopathy and neuropathy), increased risk of macrovascular complications (ischaemic heart disease, stroke and peripheral vascular disease), and diminished quality of life. Diabetes can broadly be categorized into two types;

type 1 diabetes (previously known as insulin-dependent or childhood-onset diabetes) is characterized by a lack of insulin production and type 2 diabetes (formerly called non-insulin-dependent or adult-onset diabetes) is caused by the body's ineffective use of insulin. It often results from excess body weight and physical inactivity.

*Complications of diabetes mellitus*²

Acute

These are diabetic ketoacidosis, non ketotic hyperosmolar state (NKHs). Both disorders are

associated with absolute or relative insulin deficiency, volume depletion, altered mental status. Both are potentially serious complications if not promptly diagnosed and treated.

Chronic

These are microvascular (retinopathy, neuropathy and nephropathy) and macrovascular (coronary artery disease, peripheral vascular disease and cerebrovascular disease).

Diabetic retinopathy may begin to develop as early as 7 years before the diagnosis of diabetes in patients with type 2 diabetes.³ Diabetic neuropathy is recognized by the American Diabetes Association (ADA) as “the presence of symptoms and/or signs of peripheral nerve dysfunction in people with diabetes after the exclusion of other causes.”⁴ As with other microvascular complications, risk of developing diabetic neuropathy is proportional to both the magnitude and duration of hyperglycemia. More than 80% of amputations occur after foot ulceration or injury, which can result from diabetic neuropathy.⁵ The precise nature of injury to the peripheral nerves from hyperglycemia is not known but likely is related to mechanisms such as polyol accumulation, injury from AGEs, and oxidative stress. Diabetic nephropathy is one of the major complication of both Type-1 and 2 diabetes mellitus.⁶ The classical definition of diabetic nephropathy is a progressive rise in urine albumin excretion, coupled with increasing blood pressure leading to declining glomerular filtration and eventually end stage kidney disease. These patients generally have diabetic retinopathy also.⁷ Microalbuminuria, which is defined as 30-300 mg/day in 24 hours collection or 30 - 300 micro/mg creatinine in spot collection. It is an important predictor for progression of overt proteinuria i.e. > 300mg/day. Once overt nephropathy occurs pathological changes become irreversible.⁸

Although good glycemic control reduces the incidence of microvascular complications, retinopathy and nephropathy progress in some patients, despite optimal metabolic control, suggesting that factors other than glycemia such as abnormalities in both lipid and hemostatic parameters, may play a role in the development of diabetic microangiopathy.⁹⁻¹² Thus, an altered lipoprotein profile leading to a pro-atherogenic pattern has been described in diabetes mellitus and a hypercoagulable state may be an aggravating factor.¹³

In diabetic patients, a pro-coagulant state is observed, which could contribute to the risk of catastrophic cardiovascular events. The plasma levels of many clotting factors including fibrinogen, factors VII, IX, XII, kallikrein and vWF are elevated in diabetes.¹⁴⁻¹⁶ This hypercoagulable state could be caused by an imbalance between haemostatic factors in plasma, and the endothelial cell surface. So far, indirect evidence for a state favouring thrombin formation has been gathered, mainly using plasma parameters like fibrinogen and

antithrombin activity or endothelium-dependent parameters like vWF.^{17,18}

The aim of our study is to determine distribution of plasma fibrinogen levels in type 2 diabetic patients, to study the association of fibrinogen level with HbA1c and albumin excretion rate, and to assess the relationship of fibrinogen levels with development of microvascular complications.

METHODS

In this study total 90 subjects were enrolled, out of which 60 were diabetic patients (cases) and 30 were age and sex matched non-diabetic healthy controls. 60 diabetic patients were divided into two groups of 30 each.

Group A: Patients with diabetes mellitus with microvascular complications (n = 30).

Group B: Patients with diabetes mellitus without microvascular complications (n = 30).

Controls were assigned as group C (n = 30).

Patients with diabetes mellitus with microvascular complications (Group A) were further divided into three sub-groups: Group A1 - diabetes mellitus with nephropathy; Group A2 - diabetes mellitus with retinopathy and Group A3 - diabetes mellitus with neuropathy.

According to the ADA criteria for the Diagnosis of Diabetes Mellitus, patients were assigned a diagnosis of diabetes mellitus if:¹⁹

- Symptoms of diabetes plus random blood glucose concentration >11.1 mmol/L (200 mg/dL) or
- Fasting plasma glucose >7.0 mmol/L (126 mg/dL) or
- HBA1C >6.5% or
- Two-hour plasma glucose >11.1 mmol/L (200 mg/dL) during an oral glucose tolerance test

Relevant history was taken, detailed clinical examination was done in all patients. All the patients underwent the following routine investigations.

- FBS, PPBS.
- Complete blood count.
- Renal function tests, including electrolytes.
- Glycosylated haemoglobin (HBA1C)
- Fasting Lipid profile
- Urine routine and microscopy study.
- Chest radiography.

In addition serum fibrinogen levels was also done. For serum fibrinogen levels 7 ml blood sample was collected in special tubes containing 3.2% sodium citrate. Serum

fibrinogen levels were done by turbidometric immunoassay.²⁰ Microvascular changes were assessed in all the cases. Retinopathy was diagnosed on the basis of fundus examination. Patient with microalbuminuria or overt proteinuria were considered to have nephropathy, which was calculated by urine albumin creatinine ratio (UACR).²¹

The diagnosis of neuropathy was based on presence of symptoms and signs of neuropathy i.e. any or combination of neuropathic pain, distal sensory loss, motor weakness or isolated cranial nerve palsies, and autonomic symptoms such as orthostatic hypotension, abdominal bloating, constipation, diarrhea, erectile dysfunction and biothesiometry.²²

Exclusion criteria

- Subjects with clinical/laboratory signs of liver cirrhosis

- Malignancy
- Coagulation disorder
- CAD
- CVA
- Peripheral vascular disease
- Patients on anticoagulants, antiplatelet agents, oral contraceptives, hypolipidemic drugs.

Comparisons were drawn between test groups and control group and within the test groups. Data collected was statistically analyzed.

RESULTS

Mean age among, diabetics with microvascular complications was 58.37±13.24 years, diabetics without microvascular complications was 53.80±10.36 years and controls was 53.37±7.05 years (p = 0.132).

Table 1: Comparison of variables among diabetics with microvascular complications (Group A), diabetics without microvascular complications (Group B) and controls (Group C).

Variables	Cases (Group A, n = 30)	Cases (Group B, n = 30)	Controls (Group C, n = 30)	P-value
Age (years)±SD	58.37±13.24	53.80±10.36	53.37±7.05	0.132
Gender (M/F)	15/15	18/12	18/12	-
Duration of diabetes (years)±SD	12.96±4.83	6.93±2.54	-	<0.001
BMI (kg/m ²)±SD	27.24±3.28	24.23±2.82	23.68±2.07	<0.001
Waist circumference (cm)±SD	87.10±6.04	82.20±6.62	83.70±4.90	0.006
Fibrinogen (mg/dl)±SD	293.43±51.09	238.90±44.12	174.66±18.32	<0.001
Platelet count (lakh/cumm)±SD	2.32±0.83	2.34±0.93	2.09±0.60	0.420
FBS (mg/dl)±SD	141.77±30.41	136.33±12.65	89.50±3.57	<0.001
HBA1C (%)±SD	9.45±2.31	8.22±2.07	4.87±0.70	<0.001
UACR (µg/mg)±SD	199.74±153.93	23.13±16.50	96.67±7.47	<0.001
Cholesterol (mg/dl)±SD	143.4±45.42	128.83±33.39	96.67±7.47	<0.001
Triglycerides (mg/dl)±SD	136.13±74.25	136.37±56.52	111±9.31	0.12
HDL (mg/dl)±SD	36.17±12.76	41.43±20.56	51.90±6.91	<0.001
LDL (mg/dl)±SD	85.50±33.59	75.40±15.16	81.97±10.00	0.204
VLDL (mg/dl)±SD	33.93±19.67	32.97±14.29	22.63±1.88	0.004

Mean age of cases (group A + B) was 56.08±12.01 years and that of controls (group C) was 53.37±7.05 years (p = 0.256). Duration of diabetes was significantly higher in diabetics with microvascular complications, group A (12.96±4.83 years) as compared to diabetics without microvascular complications, group B (6.93±2.54 years); p <0.001. Mean BMI was found to be significantly higher in diabetic cases (group A+B), 25.74±3.39 kg/m² as compared to controls (group C), 23.68±2.07 kg/m² (p = 0.003). Mean BMI among diabetic patients with microvascular complications (group A), 27.24±3.28 kg/m² was found significantly higher than diabetics without microvascular complications (group B), 24.23±2.82 kg/m² (p <0.001). Mean waist circumference

in diabetic cases (group A+B), 84.65±6.75 cm was higher than non-diabetic cases (group C), 83.70±4.90 but the difference was not statistically significant (p = 0.495). Mean waist circumference was significantly higher in group A (87.10±6.04 cm) as compared to group B (82.20±6.62 cm); p=0.004. Mean HBA1C level was found to be significantly higher in patients with diabetic microvascular complications, 9.45±2.31% as compared to diabetics without microvascular complications, 8.22±2.07% (p = 0.034). Mean UACR in group A (199.74±153.93 µg/mg) was significantly higher than group B (23.13±16.50 µg/mg); p <0.001. Difference in the level of serum cholesterol, HDL and VLDL among three groups i.e. group A, group B and group C, was

found to be statistically significant, with p-value <0.001, <0.001 and 0.004, respectively (Table 1).

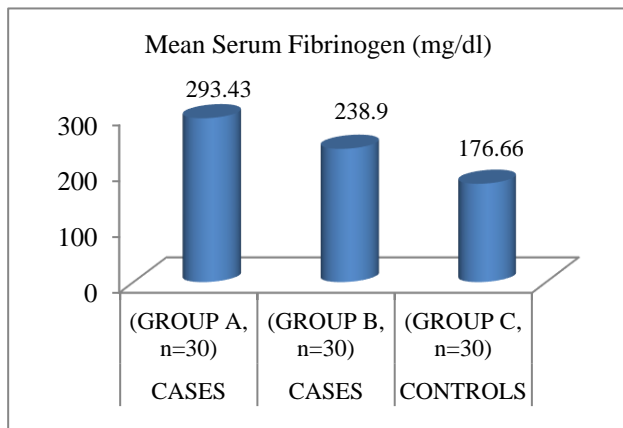


Figure 1: Comparison of mean fibrinogen levels among three study groups.

Serum fibrinogen levels were significantly higher ($p < 0.001$) in diabetic patients (266.16 ± 54.73 mg/dl) as compared to non-diabetic controls (174.66 ± 18.32 mg/dl). Further, serum fibrinogen levels were found to be significantly higher ($p < 0.001$) in diabetic patients with microvascular complications (293.43 ± 51.09 mg/dl) as compared to those without microvascular complications (238.90 ± 44.12) (Figure 1).

Diabetic patients in group A who had significantly poorer glycaemic control measured by HBA1C, $9.45 \pm 2.31\%$ as compared to group B, $8.22 \pm 2.07\%$ ($p = 0.03$) had significantly higher value of serum fibrinogen (293.43 ± 51.09 mg/dl) as compared to group B (238.9 ± 44.12 mg/dl); $p < 0.001$. Diabetic patients in group A who had significantly higher values of mean UACR, 199.74 ± 153.93 $\mu\text{g}/\text{mg}$ as compared to group B, 23.1 ± 16.50 $\mu\text{g}/\text{mg}$ ($p < 0.001$); had significantly higher serum fibrinogen levels 293.43 ± 51.09 mg/dl as compared to group B (238.9 ± 44.12 mg/dl); $p < 0.001$ (Figure 2).

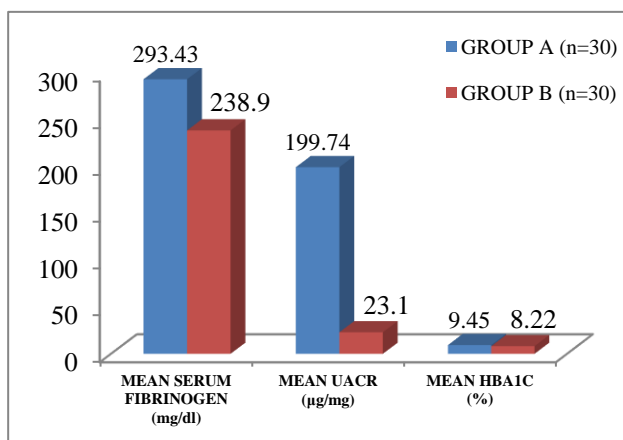


Figure 2: Comparison of mean serum fibrinogen level with UACR and HBA1C between group A and group B.

Mean serum fibrinogen level was found to be significantly higher in patients with diabetic nephropathy (301.40 ± 53.59 mg/dl) as compared to diabetics without microvascular complications ($p < 0.001$). Mean serum fibrinogen level was found to be significantly higher in patients with diabetic retinopathy (294.67 ± 32.03 mg/dl) as compared to diabetics without microvascular complications ($p < 0.001$). Mean serum fibrinogen level was found to be significantly higher in patients with diabetic neuropathy (291.42 ± 53.89 mg/dl) as compared to diabetics without microvascular complications ($p < 0.001$).

DISCUSSION

In large epidemiological studies, the procoagulant factors (fibrinogen and factor VII) have been described as independent predictors of cardiovascular events in diabetic and nondiabetic subjects. Asakawa et al. had shown that fibrinogen level was significantly higher in diabetic patients with retinopathy or nephropathy than in patients without these complications.²³ The detection of microalbuminuria, another well-known independent indicator of much greater risk of premature death from cardiovascular complications, has been demonstrated to correlate significantly with various hemostatic variables, including serum fibrinogen.^{24,25}

The present case control study was conducted at Sri Guru Ram Das Institute of Medical Sciences and Research, Amritsar, Punjab, India to study levels of serum fibrinogen in type 2 diabetes mellitus and its association with diabetic microvascular complications. In conclusion, hypercoagulable state as evidenced by increased serum fibrinogen level was responsible as one of the factors for the development of microvascular complications of diabetes mellitus. Also higher serum fibrinogen levels were positively associated with duration of diabetes, HBA1C and UACR.

Funding: No funding sources

Conflict of interest: None declared

Ethical approval: The study was approved by the institutional ethics committee

REFERENCES

- Narayan KM, Zhang P, Kanaya AM, Williams DE, Engelgau MM, Imperatore G, et al. Diabetes: the pandemic and potential solutions. In: Disease control priorities in developing countries, 2nd edition. Washington DC: World Bank. 2006:591-603.
- Powers AC. Diabetes mellitus. In: Kasper DL, Fauci AS, Hauser SL, Longo DL, Jameson JL, Loscalzo J, editors. Harrison's Principles of Internal Medicine. 19th edition. New York. McGraw Hill. 2015;419:2422-30.
- Fong DS, Aiello LP, Ferris FL, Klein R. Diabetic retinopathy. Diabetes Care. 2004;27:2540-53.

4. American Diabetes Association: Standards of medical care in diabetes. *Diabetes Care.* 2007;30:4-41.
5. Boulton AJ, Vinik AI, Arezzo JC, Bril V, Feldman EL, Freeman R, et al. Diabetic neuropathies: a statement by the American Diabetes Association. *Diabetes Care.* 2005;28:956-62.
6. Glowinska I, Grochowski J, Malyszko J. Cardiovascular complication in renal replacement therapy. *Polish Archives Internal Medicine.* 2008;118:7-8.
7. Obiniche EN, Adam A. Update in Diabetic Nephropathy. *Int J Diabetes Metabolism.* 2005;13:1-9.
8. Lin JE, Robbins DC, Bella JN, Palmieri V, Bella JN, Roman MJ. Association of albuminuria with systolic and diastolic left ventricular dysfunction in Type-2 diabetes mellitus. *J Am Cardiol.* 2003;41(11):2022-8.
9. The Diabetes Control and Complications Trial Research Group. The effect of intensive treatment of diabetes on the development and progression of long-term complications in insulin-dependent diabetes mellitus. *N Engl J Med.* 1993;329:977.
10. Klein R, Klein BEK, Moss SE. Relation of glycemic control to diabetic microvascular complications in diabetes mellitus. *Ann Intern Med.* 1996;124:90.
11. Ramsay RC, Goetz FC, Sutherland DER, Mauer SM, Robinson LL, Cantrill HL, et al. Progression of diabetic retinopathy after pancreas transplantation for insulin-dependent diabetes mellitus. *N Engl J Med.* 1988;318:208.
12. Viberti GC, Bilous RW, Mackintosh D, Bending JJ, Keen H. Long term correction of hyperglycaemia and progression of renal failure in insulin dependent diabetes. *Br Med J.* 1983;286:598.
13. Glowinska I, Grochowski J, Malyszko J. Cardiovascular complication in renal replacement therapy. *Polish Archives Internal Medicine.* 2008;118:7-8.
14. Carr ME. Diabetes mellitus: a hypercoagulable state. *J Diabetes Complications.* 2001;15:44-54.
15. Kario K, Matsuo T, Kobayashi M, Matsuo M, Sakata T, Miyata T. Activation of tissue factor-induced coagulation and endothelial cell dysfunction in non-insulin-dependent diabetic patients with microalbuminuria. *Arterioscler Thromb Vasc Biol.* 1995;15:1114-20.
16. Kvasnicka J, Skrha J, Perusicova J, Kvasnicka T, Markova M, Umlaufova A, et al. Haemostasis, cytoadhesive molecules (sE-selectin and sICAM-1) and inflammatory markers in non-insulin-dependent diabetes mellitus. *SbLek.* 1998;99:97-101.
17. Ostermann H, Loo J. Factors of the hemostatic system in diabetic patients: a survey of controlled studies. *Haemostasis.* 1986;16:386-416.
18. Carmassi F, Morale M, Puccetti R, Negri F, Monzani F, Navakesi R, et al. Coagulation and fibrinolytic system impairment in insulin dependent diabetes mellitus. *Thromb Res.* 1992;67:643-54.
19. American Diabetes Association. Diagnosis and classification of diabetes mellitus. *Diabetes Care.* 2014;37:81-90.
20. Lothar T. Clinical laboratory diagnostics. 1st Edition. TH-Books. Frankfurt, Germany. 1998:621-625.
21. Ginsberg JM, Chang BS, Matarese RA. Use of single voided urine samples to estimate quantitative proteinuria. *N Engl J Med.* 1983;309:1543.
22. Young MJ, Breddy JL, Veves A, Boulton AJM. The prediction of diabetic neuropathic foot ulcerations using vibration perception thresholds. *Diabetes Care.* 1994;17:557-60.
23. Asakawa H, Tokunaga K, Kawakami F. Elevation of fibrinogen and thrombin-antithrombin III complex levels of type 2 diabetes mellitus patients with retinopathy and nephropathy. *J Diabetes Complications.* 2000;14:121-6.
24. Knobl P, Scherthaner G, Schnack C. Thrombogenic factors are related to urinary albumin excretion rate in type 1 (insulin dependent) and type 2 (non –insulin -dependent) diabetic patients. *Diabetologia.* 1993;36:1045-50.
25. Gabazza EC, Takeya H, Deguchi H, Sumida Y, Taguchi O, Murata , et al. Protein C activation in NIDDM patients. *Diabetologia.* 1996;39:1455-61.

Cite this article as: Mohan G, Kaur R, Aggarwal A, Singh P. To study levels of serum fibrinogen in type 2 diabetes mellitus and its association with diabetic microvascular complications. *Int J Adv Med* 2017;4:10-4.