ORIGINAL ARTICLE

# CORRELATION OF PLASMA FIBRINOGEN LEVELS WITH VARIABLES IN PATIENTS OF TYPE-II DIABETES MELLITUS WITH MICROVASCULAR COMPLICATIONS

***Imran Ali Zaidi[1], Anila Jaleel[2], Kiran Namoos[1], Hassan Ali[3], Amna Iqtidar[4], Farouk Qamar Malik[1]***

*[1]Department of Biochemistry, Fatima Memorial College of Medicine and Dentistry, Lahore, [2] Department of Biochemistry. Shalamar Medical College Lahore, [3]Department of Biochemistry, Institute of Molecular Biology and Biotechnology, The University of Lahore, [4]Department of Paediatric Medicine, Unit-II Services Hospital, Lahore.*

**ABSTRACT**

**Background:** Diabetes mellitus is considered as hypercoagulable state, where hypercoagulability of blood result in acceleration of atherosclerosis and diabetic microvascular complications. Three recognized diabetic microvascular complications include diabetic neuropathy, diabetic nephropathy and diabetic retinopathy. The aim of the present study was to evaluate association of plasma fibrinogen levels with other variables in patients with any of the three-recognized diabetic microvascular complications: diabetic neuropathy, diabetic retinopathy or diabetic nephropathy.

**Methods:** 104 patients of T2DM from in Medical unit-II and ophthalmology ward, Services Hospital, Lahore from April to October 2017 were included in present study. The patients were divided into two groups of 52 patients each. Group 1 comprised of Type 2 Diabetes Mellitus patients without any diabetic microvascular complications and Group II: Type 2 Diabetes Mellitus patients with any of three recognized diabetic microvascular complications: diabetic neuropathy, diabetic nephropathy and diabetic retinopathy (52 patients). Plasma fibrinogen levels, blood sugar fasting (BSF), HbA1c and BMI were evaluated in all patients.

**Results:** Spearman's rank correlation coefficient, applied for measuring correlation of variables, showed statistically significant correlation ($P < 0.05$) of BSF $r = 0.39$; HbA1c $r = 0.48$ to plasma fibrinogen in patients with any of the three diabetic microvascular complications.

**Conclusion:** Higher fibrinogen levels (Clotting factor I) in plasma contribute significantly to establishment of microvascular complications in type-II diabetes mellitus patients. Fibrinogen levels were positively correlated with HbA1c and blood sugar fasting.

**Key words:** Plasma fibrinogen, microvascular complications, diabetes mellitus.

***Corresponding Author:***
Dr. Imran Ali Zaidi,
Department of Biochemistry,
Fatima Memorial College of Medicine and Dentistry,
Lahore.
E-mail: imran-sir@hotmail.com

## INTRODUCTION

Diabetes mellitus (DM) is defined as a metabolic disorder due to multiple etiological factors and is featured by chronic hyperglycemia amalgamated with derangements of carbohydrate, protein and fat metabolisms causing long–term morbidity, dysfunction and failure of multiple body organs.[1]

Type II diabetes mellitus (T2DM), due to gradual loss of pancreatic β-Cell function, results in impaired secretion of insulin and increased requirement for insulin due to insulin resistance. Microvascular complications of DM include diabetic neuropathy, diabetic nephropathy and diabetic retinopathy, which are conditions manifesting from prolonged excess glucose levels in the small blood vessels supplying various body tissues[2-5].

The pathophysiology of vascular complications

include hypercoagulable state and disturbed coagulation cascade in diabetes leading to extensive platelet aggregation and hence more clot formation.[6] Many factors can affect plasma fibrinogen levels. It increases with an advancing age, body mass index (BMI), diabetes mellitus, tobacco smoking and in post-menopausal women. It declines with regular physical exercise, moderate alcohol consumption, high density lipoprotein cholesterol (HDL cholesterol) and with synthetic hormone therapy.[7, 8]

Platelet aggregation seems to execute a major role in pathophysiology of hemostasis and thrombosis. This aggregation takes place through the interface of plasma fibrinogen with its receptor on platelets which is Glycoprotein IIb-IIIa complex. Fibrinogen is an active participant in a number of pathophysiological processes in the body including inflammation, thrombogenesis and atherogenesis.[9] In the past few years, variation in hematological indices has been related to development of microvascular complications in diabetics. Few hematological factors such as blood viscosity and others are thought to participate significantly for microvascular complications in diabetics.[10]

The aim of our study was to evaluate an association of plasma fibrinogen with blood levels of glycated hemoglobin (HbA1c), fasting glucose and BMI in patients having any of the three diabetic microvascular complications.

## METHODS

This Cross-sectional study was conducted in Medical unit-II and Ophthalmology ward, Services Hospital, Lahore and in the Institute of Molecular Biology and Biotechnology (IMBB), The University of Lahore from 15th April 2017 to 15th October 2017. Informed consent was taken from all patients recruited and their demographic data was recorded. The clinical parameters were also noted.

The study included one hundred and four patients of Type-II Diabetes Mellitus with an age range 20-75 males and females.. These patients were divided into two groups for comparison; Group I: Type-II Diabetes Mellitus (T2DM) patients without any microvascular complication (52 patients). Group II: Type 2 Diabetes Mellitus patients with any of the three recognized microvascular complications: Diabetic neuropathy, diabeticretinopathy or diabetic nephropathy (52 patients). Patients with any systemic illness altering the blood coagulability, inherited bleeding disorders, severe liver or cardiac failure or cancer, on oral contraceptive pills or who underwent surgical operation in last six months, or genetic disorders like hemoglobinopathies were excluded.

Hematological and biochemical analysis were done by using Amax destiny plus fully automated coagulation analyzer by Trinity Biotech. Hemoglobin A1c (HbA1c), fasting plasma glucose were measured on chemistry analyzer (Hitachi 902fully automated analyzer) manufactured by Roche Diagnostics Germany. Body mass index (BMI) was done by using the formula: weight measured in kilograms divided by height measured in square meter (kg/m2).[11]

The statistical analysis of the data was done by using Statistical Package for the Social Sciences; SPSS version 21 for windows (SPSS Inc., Chicago, IL, USA). Descriptive results were expressed as: mean ± SD (standard deviation). An independent samples t-test was utilized to compare means of variables. Spearman's rank correlation coefficient was utilized for finding correlation between plasma fibrinogen levels and other clinical and laboratory parameters. P values $< 0.05$ were considered statistically significant.

## RESULTS

In our study, 104 T2DM patients were registered and were divided in two groups for comparison:
The mean age of the patients in Group I was 56.02 ± 8.52 years and mean age of the patients in Group II was 55.12 ± 9.91 years. The mean blood sugar fasting in Group I was 133.71 ± 29.80 and mean blood sugar fasting in Group II was 159.12 ± 37.12. Mean HBA1c in Group I was 6.73 ± 1.25 and mean HBA1c in Group II was 8.13± 1.50. Mean plasma fibrinogen in Group I was 295.98 ± 59.70 and mean fibrinogen level in Group II was 351 ± 62.78 .Mean BMI in Group I was 23.70 ± 3.96 and mean BMI in Group II was 22.98 ± 4.15 (Table 1).

An independent samples t-test revealed that there was a significant difference between the two groups statistically as regards plasma fibrinogen levels, blood sugar fasting and HbA1c (Table 1). According to Spearman's rank correlation coefficient, following variables were statistically significantly correlated ($P < 0.05$) to plasma fibrinogen levels: Blood sugar fasting $r = 0.59$; HbA1c $r = 0.79$ (Table 2).

A bivariate Spearman's rank correlation coefficient was used to examine whether there was a relationship between diabetic microvascular complications and plasma fibrinogen. Using a two-tailed 0.05 criterion, the results revealed a significant and positive relationship between the two variables [$r (104) = 0.46$, $p < 0.05$]. The correlation was moderate in strength. Higher levels of plasma fibrinogen were associated with diabetic microvascular complications (Table 3).

**Table 1: Age, BMI, Plasma Fibrinogen, Blood Glucose and HbA1c in patients with and without microvascular complications. The values are shown as mean ±Standard error of mean.**

| TEST NAME | T2DM WITH MVC (n=52) | T2DM WITHOUT MVC (n=52) | P VALUE |
|---|---|---|---|
| **Age (years)** | 56.02± 1.18 | 55.12± 1.37 | 0.62 |
| **Body mass index (BMI)** | 22.98±0.57 | 23.70±0.54 | 0.37 |
| **Plasma fibrinogen** | 351.08±8.70 | 295.98±8.27 | .000 |
| **Blood sugar fasting** | 159.12±5.14 | 133.71±4.13 | .000 |
| **HBA1c** | 8.13±0.20 | 6.73±0.17 | .000 |

**Independent samples T test: P < 0.05: Statistically significant**

**Table 2: Relationship between plasma fibrinogen and variables.**

| Tested variables | N | Spearman's rho correlation coefficient(r) | P value |
|---|---|---|---|
| **Blood sugar fasting** | 104 | .587 | .000 |
| **HBA1c** | 104 | .785 | .000 |
| **BMI** | 104 | .048 | .628 |
| **Age** | 104 | -.021 | 0.83 |

**Statistical analysis by Spearman's rank correlation coefficient**

**Table 3: Relationship between diabetic microvascular complications and plasma fibrinogen. Statistical analysis by bivariate Spearman's rank correlation coefficient.**

| | | | Groups | Fibrinogen |
|---|---|---|---|---|
| Spearman's rho | Groups | Correlation Coefficient | 1.000 | .458** |
| | | Sig. (2-tailed) | . | .000 |
| | | N | 104 | 104 |
| | Fibrinogen | Correlation Coefficient | .458** | 1.000 |
| | | Sig. (2-tailed) | .000 | . |
| | | N | 104 | 104 |

## DISCUSSION

Diabetes Mellitus is one of the major causes of morbidity not only in developed but also in developing countries. Blood hypercoagulability in diabetes mellitus may result in acceleration of atherosclerosis and development of diabetic microvascular complications.[12]

One of the contributing factor to increased blood viscosity and hypercoagulability is elevated plasma fibrinogen levels which is considered as a cardinal risk factor for development of diabetic microvascular complications.[13]

Our study shows significantly elevated plasma fibrinogen level in T2DM patients with diabetic microvascular complications i.e. diabetic neuropathy, diabetic retinopathy or diabetic nephropathy as compared to T2DM patients without any diabetic microvascular complication. The findings are consistent with study done by Mohan et al. that showed significantly increased serum fibrinogen level in 30 diabetic patients with complications as compared to controls.[14] Neetha et al. studied plasma fibrinogen levels in 61 patients with diabetic retinopathy and compared them with those without diabetic retinopathy. The study pointed out that increased plasma fibrinogen level significantly contributes to the initiation and development of diabetic retinopathy. [15]

Aslam et al. concluded significant correlation of plasma fibrinogen levels with the three microvascular complications, suggesting that hyperfibrinogenemia leads to increased vascular complications in type 2 diabetes mellitus patients.[16] Sarangi et al. point out the role of plasma fibrinogen in establishment of diabetic retinopathy.[17]

Our study showed significant correlation between plasma fibrinogen level and blood sugar fasting

and HbA1C. Significant positive correlation exists between plasma fibrinogen levels and blood sugar fasting, and plasma fibrinogen and HbA1c. However no significant correlation was found between plasma fibrinogen and body mass index in the study.

In diabetic microangiopathy there are numerous proposed phenomena explaining elevated fibrinogen level in patient of diabetes mellitus. Firstly it is thought to be linked with low grade inflammation. Exaggerated platelets, leukocytes activation and the presence of heterotypic aggregations have been documented in diabetic microangiopathy.[18] Furthermore cytokines such as Interleukin-6 levels are increased in diabetes which is believed to stimulate liver cells, that synthesizes more fibrinogen, thus suggesting a crucial connection between inflammation and hypercoagulation.[19] Insulin resistance is considered another important risk factor which leads to escalated fibrinogen hepatic production in patients of T2DM.[20]

Gupta et al. found positive correlation of glycosylated hemoglobin (HbA1c) with fibrinogen level[21]. Similarly Azad et al. found significant positive correlation of HbA1c with plasma fibrinogen level in patients with diabetic retinopathy[22].

Studies by Coban et al. and Mir et al. showed positive correlation between plasma fibrinogen level fasting blood sugar.[23,24]

In our study, non significant correlation was observed between plasma fibrinogen levels and BMI, which is in agreement with study Lyer et al.[25] but Bembde et al.[26] showed significant correlation.

This study suggests that an important role of plasma fibrinogen in development of microvascular complications in diabetes mellitus patients. However, this study had several limitations. Firstly, the number of patients enrolled was relatively small. Secondly, the study design was cross-sectional.

Additional large-scale, prospective studies are needed to further evaluate the role of plasma fibrinogen levels in development of microvascular complications in diabetic patients. It is recommended based on this study that early drug therapy and management may be initiated to lower plasma fibrinogen levels in order to prevent initiation and progression of diabetic microvascular complications.

## CONCLUSION

Higher fibrinogen levels (Clotting factor I) in plasma may contribute significantly to establishment of microvascular complications in type-II diabetes mellitus patients. Fibrinogen levels are positively correlated with HbA1c and blood sugar fasting.

## ACKNOWLEDGMENTS

The authors acknowledge Prof. Dr. M. H. Qazi for providing supervision and the working facilities for this project. The authors are also thankful to all the respected teachers of Medical unit-II, Ophthalmology ward, and Pathology department, Services hospital Lahore for their guidance and logistic support.

## REFERENCES

1. American Diabetes Association. Diagnosis and classification of diabetes mellitus. Diabetes Care 2014; 37(Suppl. 1): S81–S90.
2. Ashcroft FM, Rorsman P. Diabetes mellitus and the beta cell: The last ten years. Cell 2012; 148: 1160–71.
3. Tesfaye S, Selvarajah D: Advances in the epidemiology, pathogenesis and management of diabetic peripheral neuropathy. Diabetes Metab Res Rev 2012; 28(Suppl. 1): 8–14.
4. Haneda M, Utsunomiya K, Koya D, Babazono T, Moriya T, Makino H, et al. A new classification of Diabetic Nephropathy 2014: a report from Joint Committee on Diabetic Nephropathy.
Clin Exp Nephrol 2015; 19(1):1-5.
5. Stitt AW, Curtis TM, Chen M, Medina RJ, McKay GJ, Jenkins A, et al. The progress in understanding and treatment of diabetic retinopathy. Prog Retin Eye Res 2016; 51: 156–86.
6. Vazzana N, Ranalli P, Cuccurullo C, Davì G. Diabetes mellitus and thrombosis. Thromb Res 2012; 129(3):371-7.
7. Machlus KR, Cardenas JC, Church FC, Wolberg AS. Causal relationship between hyperfibrinogenemia, thrombosis, and resistance to thrombolysis in mice. Blood 2011; 117(18): 4953–63.
8. Mahendra JV, Kumar SD, Anuradha TS, Talikoti P, Nagaraj RS, Vishali V. Plasma Fibrinogen in Type 2 Diabetic Patients with Metabolic Syndrome and its Relation with Ischemic Heart Disease (IHD) and Retinopathy. J Clin Diagn Res 2015; 9(1): BC18-21.
9. Floyd CN, Ferro A. The platelet fibrinogen receptor: from megakaryocyte to the mortuary. JRSM Cardiovasc Dis 2012; 1(2).
10. Giacco F, Brownlee M. Oxidative stress and diabetic complications. Circ Res 2010; 107(9): 1058-70.
11. Blackburn H, Jacobs D. Commentary: Origins and evolution of body mass index (BMI): continuing saga. Int J Epidemiol 2014; 43 (3): 665–9.
12. Rask-Madsen C and King GL. Vascular complications of diabetes: mechanisms of injury and protective factors. Cell Metab 2013; 17(1): 20–33.
13. Le DS, Miles R, Savage PJ, Cornell E, Tracy RP, Knowler WC, et al. The association of plasma fibrinogen concentration with diabetic microvascular complications in young adults with early-onset of type 2 diabetes. Diabetes Res Clin

Pract 2008; 82(3): 317-23.
14. 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(1):10-14.
15. Neetha K, Vijaya P. Plasma fibrinogen levels and its relation to diabetic retinopathy. Kerala Journal of Ophthalmology 2010; Vol. XXII, No.4.
16. Aslam M, Chandrasekhara P. Correlation of Fibrinogen and HsCRP with Microvascular Complications of Type 2 Diabetes Mellitus. IJHSR 2016; 6(5): 25-32.
17. Sarangi R, Mahapatra S, Padhi S. Association of plasma fibrinogen and serum high-sensitivity C-reactive protein in type 2 diabetes mellitus. J Curr Res Sci Med 2015;1:21-6.
18. Yeom E, Byeon H, Lee SJ. Effect of diabetic duration on hemorheological properties and platelet aggregation in streptozotocin-induced diabetic rats. Sci Rep 2016; 6: 21913.
19. Carty CL, Heagerty P, Heckbert SR, Jarvik GP, Lange LA, Cushman M, et al..Fibrinogen andIL6 Gene Variants and IL-6 Levels in Relation to Plasma Fibrinogen Concentration and Cardiovascular Disease Risk in the Cardiovascular Health Study. Ann Hum Genet 2010; 74(1): 1–10.
20. Barazzoni R, Kiwanuka E, Zanetti M, Cristini M, Vettore M and Tessari P. Insulin Acutely Increases Fibrinogen Production in Individuals With Type 2 Diabetes but Not in Individuals Without Diabetes. Diabetes 2003; 52(7): 1851-6.
21. Gupta RK, Dhawale S. Association between Serum Fibrinogen Level in Type-2 Diabetes Mellitus Patient with or without Microvascular Complication. IJAR 2015; 1(9): 555-61.
22. Azad N, Agrawal L, V. Emanuele N, Klein R, D. Bahn G, McCarren M, et al and the VADT Study Group. Association of PAI-1 and Fibrinogen with Diabetic Retinopathy in the Veterans Affairs Diabetes Trial (VADT). Diabetes Care 2014; 37(2): 501–6.
23. Coban E, Sari R, Ozdogan M, Akcit F. Levels of plasma fibrinogen and dimer in patients with impaired fasting glucose. Exp Clin Endocrinol Diabetes 2005;113(1):35-7.
24. Mir AM, Rashid S, Rashid A, Maqbool R, Ahmad M, Rashid W, et al. Serum fibrinogen levels and its relation to diabetic retinopathy. JEMDS 2015; 4 (95): 16036-44.
25. Lyer MU, Desai P. Assessment of C-reactive protein and fibrinogen level in type 2 diabetes mellitus. Biomed Res 2010; 21: 208-13.
26. Bembdi AS. A study of plasma fibrinogen level in type-2 diabetes mellitus and its relation to glycemic control. Indian J Hematol Blood Transfus 2012; 28(2): 105-8.