Estimation of Plasma Fibrinogen in Type II Diabetes Mellitus and It's Correlation with Glycemic Control and Urine Albumin Excretion Rate

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ABSTRACT

Background : Patients with diabetes mellitus have 2 to 4 times increased risk for cardiovascular disease than non-diabetic patients. Fibrinogen has been identified as an independent risk factor for cardiovascular disease and it is associated with traditional cardiovascular risk factors.

Aim and Objectives : The present study was undertaken to estimate plasma fibrinogen level and its correlation with glycemic control and urine albumin excretion rate (UAER) in patients with type 2 diabetes mellitus in addition to assessment of risk factors, such as smoking, alcoholism, lipid profile, hypertension and ischemic heart disease.

Methods : Total 100 patients satisfied inclusion criteria were selected for the study, comprising 60 diabetic patients as cases and 40 non-diabetic persons as controls. Plasma fibrinogen level (Modified Clauss Method) was estimated in all cases and controls, and it was correlated with various parameters such as glycemic control and urine albumin excretion rate.

Results : Mean plasma fibrinogen level in cases was high (401.58 ± 67.62 mg/dl) as compared to controls (288 ± 64.19 mg/dl), which was found to be statistically highly significant (p=0.0000). Plasma fibrinogen was highly significant in cases as compared to controls in hypertensive patients (p=0.0000), IHD patients (p=0.0024), smokers (p=0.000) and in alcoholics (p=0.0120) and it had negative correlation with age which was statistically insignificant in both the groups. Plasma fibrinogen was higher and had positive correlation with increased duration of diabetes (p=0.0000) and was significantly raised with increased UAER and with rising levels of glycosylated Hb. On multivariate analysis, both glycosylated Hb(P=0.004) and UAER(P=0.038) were independently correlated with fibrinogen irrespective of other confounding factors.

Conclusion : Patients with type 2 diabetes mellitus had a higher plasma fibrinogen level. Fibrinogen level was significantly correlated with glycemic control and urine albumin excretion rate independent of other risk factors.

Keywords : Fibrinogen level, Glycemic control, Urine albumin excretion rate, Diabetes mellitus, Risk factors.

Introduction :

Diabetes mellitus is the most common metabolic disorder characterized by metabolic abnormalities and long term complications. The chronic complications of diabetes mellitus affect many organ systems and are responsible for the majority of morbidity and mortality associated with the disease[1]. The prevalence of both type 1 and type 2 diabetes mellitus is increasing worldwide, but the prevalence of type 2 diabetes mellitus is expected to rise more rapidly in future because of increasing obesity and reduced activity levels[2].

In the past decade, the potential role of hemostatic factors, particularly fibrinogen, in atherosclerosis and its complications has generated considerable attention. Increasing evidence from epidemiological studies suggest that elevated plasma fibrinogen levels are associated with an increased risk of cardiovascular disorders, especially in diabetic patients[3,4]. Also the plasma concentration of fibrinogen predicts cardiovascular events in both the general population and non-diabetic patients with clinical vascular disease. The excess cardiovascular morbidity and mortality among diabetics have not been fully explained by major risk factors such as hypertension, cigarette smoking and hypercholesterolemia. Increased attention is being

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paid to, disordered hemostatic mechanism in pathogenesis of both large vessel and small vessel disease in diabetes[5].

The increase in urine albumin excretion rate is a marker of poor control of diabetes. Microalbuminuria has been recognized as an important biomarker to predict micro vascular and macrovascular diabetic complications[6]. Poor glycemic control has been reported to be associated with increased vascular complications in diabetics[4]. A handful studies have been done regarding the association of fibrinogen with glycemic control and urine albumin excretion rate in type 2 diabetes mellitus. Therefore, we planned to study plasma fibrinogen level in patients with type 2 diabetes mellitus and its correlation with glycemic control and urine albumin excretion rate in addition to assessment of risk factors, such as smoking, alcoholism, lipid profile, hypertension and ischemic heart disease.

Materials and Methods :

The present case control study was carried out in total 100 (n=100) patients of either sex, aged 34-60 years, irrespective of history of hypertension, ischemic heart disease (more than 1 month duration), cerebrovascular episode, smoking, alcoholism, tobacco chewing etc either indoor in medicine wards or attendees of diabetic clinic, Government Medical College and Hospital, Nagpur for a period of 12 months. 60 cases (n=60) were patients with type 2 diabetes mellitus with or without microvascular complications and 40 subjects were (n=40) non-diabetic age, sex, and body mass index matched controls irrespective of history of hypertension, IHD, cerebrovascular episode, smoking, alchohol, tobacco chewing. Exclusion criteria were patients with type 1 DM, age > 60 yrs, pregnancy, nephrotic syndrome, Acute MI(<4weeks), associated inflammatory or neoplastic conditions, CCF, women on OCP. Institutional Ethical Committee approval was taken. Patients with type 1 diabetes mellitus, having age more than 60 years, pregnancy, nephritic syndrome, acute myocardial infarction (within 4 weeks), associated inflammatory or neoplastic conditions, congestive cardiac failure, women on oral contraceptive drugs were excluded from the study.

A detailed history and clinical examination was done pertaining to various risk factors, and relevant laboratory investigations were done in both diabetic patients and in controls. The various parameters that were studied included age of the patient (year), sex, BMI (kg/m2), history of smoking, alcoholism, hypertension and Ischemic heart disease (IHD). The blood sugar level, lipid profile, plasma fibrinogen levels (mg/dl) by "Modified Clauss method" and glycosylated hemoglobin (%) by "cation exchange resin method". Urine albumin was estimated from quantitative albuminuria by immune electrophoresis and UAER was calculated in cases and controls.

Statistical Analysis :

Statistical analysis was done by using student's test for continuous variables and chi square test for categorical variables. All these tests along with multilogistic regression analysis were done by using software Stata version 7.0 © Stata Corporation 1997-2000. P value of □0.05 was considered significant.

Observations and Results :

The mean age of patients included in the study was 51.63 ± 7.09 with range of 35 to 60 years in cases and mean age in controls was 50.13 ± 6.96 with range of 34 to 60 years. The majority of cases (58.33%) and controls (60%) were in age group of 51-60 years. Maximum patients (60%) of both the groups had grade II obesity (BMI □30-40 kg/m^2) and most of the cases (58.33%) had duration of diabetes more than 10 years. In cases, there were 51.66% males and 48.33% females while in controls 52.5% were males and 47.5% were females.

Table 1 shows the descriptive analysis of parameters including conventional risk factors and laboratory investigations. Also table shows that the percentage of risk factors was more in control group than cases. None of the females among cases or control group were smoker or alcoholic.

Table 1 : **Descriptive analysis in diabetes and controls**

Parameters	Cases	Controls	P- value
Age	51.63 ± 7.08	50.13 ± 6.93	0.2974
Duration of DM (years)	10.15 ± 4.4	-	
Hypertension	22 (36.66%)	16 (40%)	0.4432
IHD	06 (10%)	12 (30%)	0.3452
Smoking	16 (26.66%)	11 (27.05%)	0.5288
Alcohol	08 (13.33%)	08 (20%)	0.3740
Mean blood Sugar Fasting (mg/dl)	166.0 ± 62.82	79.55 ± 14.68	<0.001
Mean blood Sugar Postmeal (mg/dl)	264.38 ± 95.85	-	
Mean glycosylated Hb (%)	8.37 ± 1.87	4.62 ± 0.82	<0.001
Mean UAER (µg/min)	49.346 ± 23.52	15.76 ± 17.6	0.000
Serum Cholesterol (mg/dl)	219.48 ± 49.71	190.4 ± 47.03	<0.001
Serum TGL (mg/dl)	187 ± 53.72	168.12 ± 38.06	<0.001
Fibrinogen (mg/dl)	401.58 ± 67.62	288 ± 64.19	0.000

Mean plasma fibrinogen level was raised in statistically significant manner in diabetics (401.58 ± 67.62 mg/dl) than controls (288 ± 64.19 mg/dl), (p=0.0000), ***(Table 2).***

Table 2 : **Distribution of patients according serum fibrinogen levels**

Serum Fibrinogen	Cases	Controls
≤350	07 (11.66%)	26 (65%)
□350-400	15 (25%)	13 (32.5%)
□ 400-450	17 (28.33%)	01 (2.5%)
□ 450	21 (35%)	00

Mean urine albumin excretion rate (UAER) in cases and controls was 49.346 ± 23.52 (µg/min) and 15.76 ± 17.6 (µg/min) respectively, it was significantly higher in diabetics than controls (p=0.0001), ***(Figure 1).***

Figure 1 : **Distribution of plasma fibrinogen in cases and controls in relation to UAER**

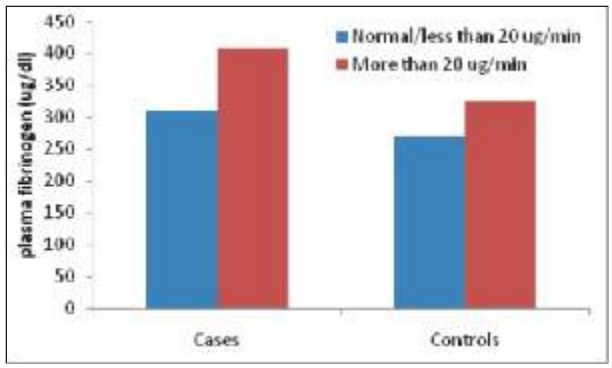

Table 3 shows the distribution of plasma fibrinogen in relation to various risk factors.

Table 3 : **Distribution of plasma fibrinogen in cases and controls**

Parameters	Groups	Plasma Fibrinogen		P value
		Cases	Controls	
Age	34-40	426.42 ± 54.97	363.75 ± 11.81	<0.001
	41-50	422.23 ± 60.71	302.5 ± 68.8	<0.001
	51-60	386 ± 70.40	268.125 ± 56.45	<0.001
Sex	Male	397.42 ± 71.56	307.36 ± 68.56	<0.001
	Female	406.03 ± 56.11	266.57 ± 52.75	<0.001
Duration of Diabetes (Years)	≤5	295 ± 55.37	-	
	□5-10	376.667 ± 50.32	-	
	□ 10	435.71 ± 47.56	-	

Hypertension	Present	443.63 ± 52.08	320.31 ± 63.15	<0.001
	Absent	377.23 ± 63.97	266.45 ± 56.36	
IHD	Present	438.34 ± 56	324.16 ± 66	<0.001
	Absent	397.5 ± 68	272.5 ± 57.5	
Smoker	Present	455.31 ± 28.19	315.9 ± 73.47	<0.001
	Absent	382.04 ± 67.32	277.41 ± 58.22	
Alcohol	Present	370.625 ± 51.92	289.37 ± 67.44	<0.001
	Absent	406.34 ± 68.89	287.65 ± 66.02	
Glycosylated Hb (%)	≤6	-	288 ± 64.19	<0.001
	6-7	344.54 ± 47.24	-	
	7-8	372.4 ± 56.42	-	
	□ 8-9	423.34 ± 37.86	-	
	□ 9-10	440 ± 39.5	-	
	□ 10	484.09 ± 24.57	-	
UAER (µg/min)	≤20	310 ± 54.42	272.14 ± 57.75	<0.001
	□ 20	409.9 ± 62.69	325 ± 65.5	0.000

Table 4 shows multivariate analysis of correlation of plasma fibrinogen with UAER and glycosylated Hb. This analysis shows that both glycosylated Hb and UAER were significantly raised in diabetes independent of presence of other risk factors.

Table 4 : **Showing multivariate analysis**

	Odds Ratio	**Pvalue**	**95% Conf. Interval**
UAER	1.028	0.038	1.001552
Glycosylated Hb	1.66	0.004	1.17194

Discussion :

Increased cardiovascular morbidity and mortality in diabetic patients is very well understood, but underscores the need to identify potential reversible cardiovascular risk factors in this group of patients. For many years, hemostatic factors especially fibrinogen[2], has been implicated as a cause of atherosclerosis and its complications i.e., MI, angina etc irrespective of presence of diabetes mellitus. Plasma fibrinogen is an inflammatory marker and has an important role in pathogenesis of inflammation, atherosclerosis, thrombogenesis and development of vascular complications in type-2 diabetes mellitus patients[1].

The mechanisms by which fibrinogen increases cardiovascular risks are not fully understood. Fibrinogen plays important role in development of atherosclerosis starting from the stage of plaque formation till formation of occlusive thrombus over a ruptured atherosclerotic plaque, which is the most common precipitating cause of MI. The various mechanism by which fibrinogen has been found to promote atherosclerosis and thrombosis are (a) hyperfibrinogenemia increases plasma viscosity, (b) it binds its receptors on platelet membrane leading to platelet aggregation, (c) it induces reversible RBC aggregation, (d) it forms fibrin and fibrinogen degradation products (FDPs) which in turn bind LDL and sequester more fibrinogen and (e) fibrinogen and FDPs stimulate smooth cell proliferation and migration[2,7].

In our study, patients with DM had significantly higher fibrinogen level than controls. Similar results were obtained by previous studies[1,3,4,8,9]. Plasma fibrinogen was found to be decreased as the age advanced in both cases and controls which were showing negative correlation in both the groups, this was statistically insignificant. It was more in females than males in cases but difference was statistically insignificant while it was more in males

than females in controls and difference was statistically significant. This difference in fibrinogen level between males and females might be due to more risk factors in male patients from control group. But the level of plasma fibrinogen in both males and females was more in cases as compared to controls; this difference may suggest that diabetes is itself responsible for rise in fibrinogen level.

In the present study, higher levels of fibrinogen were seen in patient with hypertension and IHD compared to without hypertension and IHD; however, difference in fibrinogen among cases and controls in those with hypertension was statistically highly significant and in those with IHD was statistically significant. The fibrinogen level was significantly raised in smokers with diabetes than smokers with non diabetics. This may suggest that diabetes itself was responsible for this rise in fibrinogen. There was statistically significant difference in fibrinogen levels between cases and controls with alcoholism, even though it was decreased in alcoholics with diabetics. Our findings were compared with other studies[10-15].

Plasma fibrinogen was found to be increased as the duration of diabetes mellitus and glycosylated Hb level increased and this rise was statistically highly significant and showed positive correlation. These results were similar to previous studies[2,3,16-19]. The correlation between glycemic control and fibrinogen levels could be due to (a) glycosylate fibrinogen is less susceptible to plasmin degradation (b) relative insulin deficiency in diabetic□s results in differential protein synthesis i.e., 29% decrease in albumin synthesis and 50% increase in fibrinogen synthesis[19]. Thus, a link between fibrinogen and atherosclerosis is undeniable; it is the nature of association that is debatable-risk factor or risk marker. The difference of UAER between cases and controls was statistically highly significant (p=0.000) but serum fibrinogen levels difference with UAER□20 µg/min in cases and controls was statistically significant (0.0000). Our study correlated with different studies[17,18,20,21]. The multivariate analysis showed that plasma fibrinogen was significantly correlated with rise in glycosylated Hb and rise in urine albumin excretion rate independent of other confounding factors. This was compared with study of Saini et al[16] and Jain et al[17].

Our observation supports the hypothesis that inflammation and impaired fibrinolysis may play a pathogenetic role in T2DM and its associated micro and macrovascular complications. Strong association between fibrinogen and glycemic status also points to the fact that fibrinogen may be considered as an inflammatory marker in T2DM. Understanding the dysglycemia-inflammation coagulation paradigm may be useful in lowering the future cardiovascular risk in T2DM.

All patients with hyperfibrinogenemia should need follow up with treatment for outcome like IHD and whether decreased levels of fibrinogen either by life style modification or drugs decreases risk of IHD etc. should be looked for. We did not study effect of any drugs on fibrinogen levels and no newer risk factors were studied.

Limitations of Study :

1. This study had small sample size. Findings therefore need to be confirmed in a large study.
2. In the study Clauss Method as a functional assessment of clottable fibrinogen was used. Whether quantitative protein assay (gravimetric, immunonephalometric) or a combination of functional and quantitative assays provide accurate assessment of risk remains to be undefined.
3. High risk factors like smoking and alcohol were more in males both in cases and control group. Hence result may be varied and correlation could not be done in male and female.

Conclusion :

Plasma fibrinogen was significantly raised in diabetic patients than non-diabetic controls irrespective of presence of various confounding factors like hypertension, IHD, smoking, alcohol. The increase in urine albumin excretion rate as well as increase in level of glycosylated Hb which is a marker of poor control of diabetes showed increased levels of plasma fibrinogen significantly

independent of other confounding factors. It can be concluded that hyperfibrinogenemia may precede the onset of clinical vascular complications. This may suggests that simple routine lab investigations in DM along with easily measurable marker of inflammation and hypercoagulability i.e. plasma fibrinogen can be used in identifying patients who are at high risk for development of vascular complications in DM (Type 2).

Good control of diabetes will reduce fibrinogen levels leading to decreased complications of diabetes. Further larger studies are needed to study the plasma fibrinogen level in diabetic patients with microvascular complications and effect of interventions done to decrease the fibrinogen levels and cardiovascular mortality.

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