EUROPEAN ACADEMIC RESEARCH
Vol. IV, Issue 1/ April 2016

Impact Factor: 3.4546 (UIF)
DRJI Value: 5.9 (B+)

ISSN 2286-4822
www.euacademic.org

Fibrinogen level in Sudanese diabetic patients with macro and micro vascular complication

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Abstract:

Objective *—To evaluate the determinants of fibrinogen level in sudanese diabetes and impact of hyperfibrinogenemia on macro- and microvascular complication*

Research Design And Methods*—Plasma fibrinogen, glucose, HbA*$_{1c}$ *and lipids were measured in 120 ambulatory type I and type II diabetic patients with microvascular complication (n = 60) classified as retinopathy (n = 30), nephropathy (n =30) and macrovascular (n = 30) both depend on the clinical evidence and all compared with normal diabetes as control (n=30).*

Results*—Overall mean ± SD fibrinogen levels in patients (354.7 ±40.59mg/dl) were elevated markedly compared with control subjects (167.6±17.47mg/dl). Fibrinogen levels were elevated proportionately in patients with type II diabetes (P<0.000), hypertension (P<0.005), and with vascular complications (P<0.000). Fibrinogen was correlated significantly with age (P<0.000), Duration (p<0.000), plasma glucose (P<0.000) and with cholesterol and LDL (p<0.000), but not with triglycerides and HDL, Stepwise multiple regression analyses revealed that type II diabetes and presence of vascular complications were major determinants of fibrinogen. For*

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vascular complications, fibrinogen emerged as one of only three independent predictors, the other two being diabetes duration and hypertension.

Conclusions— *Fibrinogen frequently is elevated in diabetes and is an independent predictor of vascular complication*

Key words: fibrinogen levels, HBA_1C, HDL, LDL.

INTRODUCTION

Prospective, epidemiological studies from Goteborg, Sweden (1), London, UK (2), and Framingham, MA (3), have identified elevated fibrinogen as a risk factor for cardiovascular disease. This relationship persisted in multivariate analyses, taking into account traditional risk factors, which included smoking, cholesterol, and hypertension. In the Northwick Park Heart Study (2), homeostatic factors, i.e., fibrinogen and factor VII coagulant activity were stronger predictors for ischemic heart disease than was cholesterol. In another long-term prospective study (4), fibrinogen levels were a strong and independent predictor of acute heart attacks after adjusting for systolic blood pressure and cholesterol. Along with evidence for a role of fibrin deposition in the development of atherosclerotic lesions (5,6), such observations provide support for the theory of thrombogenesis in the evolution of atherosclerosis[7].Abundant evidence has accumulated to suggest that atherosclerosis is accelerated in both type I (8,9) and type II (10,11) diabetes. Traditional risk factors (hyperlipidemia, hypertension, smoking, age, obesity) do not account fully for the increased prevalence and severity of vascular disease in diabetes (12). It has been proposed that a hypercoagulable state in diabetes may contribute at least in part (13). Of the various hematological factors, elevated fibrinogen as a risk factor in diabetes has received little attention. A few cross-sectional studies have indicated a state of hyperfibrinogenemia in diabetes compared

with nondiabetic control subjects (14-18), particularly in those with preexisting micro- or macrovascular complications. However, these studies were not controlled for confounding variables. In this study, we examined the relationship of plasma fibrinogen and other clinical variables to vascular complications in 90 diabetic patients with a wide range of diabetes duration, severity, and glycemic control compared with 30 diabetes non complicated as controls.

RESEARCH DESIGN AND METHODS

The study population included 120 diabetic outpatients of both diabetic control and complicated cases presenting for an office visit on Sudan cardiac centre , Al nahar eye hospital and Al shheeda Selma haemodialysis centre over a period of several weeks. Pertinent information, including the presence or absence of clinical evidence of vascular complications in each patient, was recorded by the same researcher. The means (and ranges) of age, diabetes duration, the sex, treatment modalities, and status of vascular complications are presented in Table 1.

Table (1) Study Population

	N(M/F)	Age(YR) (Range)	Duration of Diabetes(YR) (Range)	Vascular Complication			Treatment		
				None	Micro	Macro	Diet	OHA	Insulin
Control Subjects	30(10/20)	46.6±6.6 (36__58)	7.1 ±2.11 (2__12)	—	—	—	—	—	—
Patient(ALL)	90(41/49)	62.63 ± 11.61 (40__87)	13.8 ± 4.48 (5__25)	90	60	30	2	19	69
Type 1	57(41/16)	63.33±11.67 (40___87)	14. 4±4.1 (5___25)	57	38	19	0	0	57
Type2	33 (0/33)	61.42±11.59 (46___82)	13.36±5. 1 (5___24)	33	22	11	2	19	12

Values for age and Duration of Diabetes are Mean ±SD, Type1 and Type 2 patients had both Micro and Macro Vascular Complications

Microvascular complications included presence of background or proliferative retinopathy and/or nephropathy. Retinopathy was classified based on the funduscopic and fluorescein

angiographic assessment, Nephropathy was defined by the presence of overt, dipstick-positive proteinuria in the absence of infection or other discernible cause; most patients had also additional evidence of diabetic nephropathy. Macrovascular disease also was defined by standard clinical criteria, including a detailed checklist of history and physical examination, routine and stress electrocardiography, and noninvasive and/or invasive peripheral vascular studies in most patients.

Of the 33 patients with type II diabetes, 11 had various combinations of coronary artery disease (n = 7), peripheral vascular disease (n = 4), defined by history of lower extremity vascular bypass procedure (n = 3) or evidence of absent pulsations, and/or amputation (n = 1);). Twenty two patients, had microvascular disease. Of the 57 type I diabetes patients, 38 had microvascular disease, and 19 had macrovascular disease.

Procedures

(fasting or nonfasting) blood samples were obtained for glucose, HbA_1C , lipids (total cholesterol, triglycerides, HDL cholesterol, LDL cholesterol), and fibrinogen determinations. Plasma glucose and lipids were determined by routine autoanalyzer methodology with enzymatic techniques (19). HbA_1C was determined by an electrophoretic method (20). For plasma fibrinogen assay, the Dade thrombin clotting time methodology was used (21)

Statistical Analysis

Unpaired samples *t* tests were conducted to determine the significance of observed differences between the means of continuous and discontinuous variables. parametric analyses were applied for the normally distributed differences in patients and control subjects. Results are presented as means ± SD, Simple and multiple regression analyses using stepwise regressions were performed with SPSS version7.2.

Ethical Considerations

This project was approved by the ethical committee of the Faculty of Medical Laboratory Sciences, Alneelain University. Samples were taken with informed consent from patients, and consent also taken from the hospital administration

RESULTS

Fibrinogen levels and clinical variables:

The mean fibrinogen levels in patients as distributed by major variables, i.e., complications, hypertension, sex, and type of diabetes, are shown in Table2.

Table (2) Distribution of Fibrinogen Levels by discontinuous variables in all population

Variables	N	Fibrinogen Levels*(MG/DL)	P.Value
Complication			
No	30	167.6±17.47	<.000
Yes	90	354.7 ±40.59	<.000
Hypertension			
No	56	260.7 ±96.3	<.000
Yes	64	350 ± 53.6s	<.000
Gender			
Male	51	316 ±82.7	0.382
Female	69	301.9 ± 93.6	0.382
Type of Diabetic			
2	65	335.1 ± 76.9	<.000
1	55	275.8 ± 92.38	<.000

The table shows the mean ± SD (mini - max) and probability (P) ,T-test was used for comparison , P value ≤ 0.05 was considered significant

Mean fibrinogen levels were elevated markedly in all of the complicated diabetic patients compared with non complicated diabetic patients These differences were highly significant as determined by unpaired Student's *t* tests ($P < 0.000$). Fibrinogen levels were elevated significantly in patients with versus without hypertension, and with type II diabetes versus type I .

Table (3) Comparisons of Continuous Variables in Patients without (Group A) and With (Group B) Vascular Complications

Variable	Group A(N=30)	Group B(N=90)	P.Value
Age	46.6 ±6.6	62.63 ± 11.61	.000
Duration of Diabetes	7.1 ± 2.11	13.8 ± 4.48	.000
Plasma Glucose (GM/DL)	179.8 ± 7.08	237.5 ± 67.2	.000
HB AIC (%)	9.6 ± 1.17	10.5 ± 1.5	.008
Cholesterol (GM/DL)	148.6 ± 16.6	210±40.8	.000
Triglycerides (GM/DL)	210 ± 25	237 ± 27	0.47
HDL Cholesterol (GM/DL)	53.0 ± 2.3	50.0 ± 2.8	0.36
LDL Cholesterol (GM/DL)	119.3 ± 29.1	155.8 ± 4.43	.000
Fibrinogen (MG/DL)	167.6 ± 17.5	354 ± 40.6	.000

The table shows the mean ± SD (mini - max) and probability (P) ,T-test was used for comparison. ,P value ≤ 0.05 was considered significant

Table (3) presents a comparison of various clinical characteristics between the diabetic patients without (grouped A) and those with vascular complications (grouped B). The complicated patients were significantly older and had longer duration of diabetes. Their mean glucose levels ,HbA_1C levels ,LDL ,and mean cholesterol levels were elevated significantly, but neither triglycerides nor HDL were elevated significantly. Fibrinogen was elevated significantly

Table (4) Univariate Correlation Coefficients between Fibrinogen and continuous Variables in cases

Variable	N	Mean ± SD	R	P.Value
Age	90	58.62 ± 12.7	0.585	.000
Duration of Diabetes	90	12.13±4.97	0.621	.000
Plasma Glucose GM/DL)	90	223.1± 63.4	0.424	.000
HB AIC (%)	90	10.25± 1.49	0.190	.038
Cholesterol (GM/DL)	90	195.2±45.22	0.529	.000
Triglycerides (GM/DL)	90	223.0 ± 18.2	0.140	0.16
LDL cholesterol	90	142.4±41.6	0.315	.000
HDL Cholesterol (GM/DL)	90	51.4 ± 1.8	0.170	0.12

Univariate regressions between fibrinogen and the variables described in Table (3) are shown in Table (4). Correlations between fibrinogen and age, duration, serum glucose, cholesterol, LDL were significant, A weak but significant correlation also was observed with HbA_1C .

Stepwise Multiple Regression Analyses : In view of the potential interdependence of multiple variables such as age, duration and type of diabetes, glucose control, hypertension, and vascular complications, stepwise multiple regression analyses were performed to identify significant independent determinants for fibrinogen (Table 5) and vascular complications (Table 6) separately.

Table (5): Stepwise Correlations of Fibrinogen with Various Determinants in Multiple Regression Analyses

Parameter	Adjusted R2
Vascular Complications	0.292
Diabetes (Type 2, 1)	0.103
Vascular Complications and Type of Diabetes.	0.337
Vascular Complications and Type of Diabetes and Duration	0.453
Vascular Complications and Type of Diabetes and HBA_1C	0.354
Vascular Complications , Type of Diabetes and Duration , HBA_1C	0.460

Other parameters entered in analyses included cholesterol, triglycerides, HDL &LDL cholesterol, age, sex, glucose, type of treatment, smoking, and hypertension

The presence of vascular complications and type II diabetes contributed as major determinants of fibrinogen. Together, these two variables accounted for ~34% of the variability in fibrinogen ($r2 = 0.337$, $P < 0.005$). Duration by itself, was a relatively minor determinant ($r2 = 0.116$); however, addition of duration to vascular complication and type II diabetes raised their predictive power to ~45% ($P < 0.005$).

Table(6): stepwise correlation of presence of vascular complication with various determination

Parameter	Adjusted R2
Fibrinogen	0.289
Hypertension	0.040
Duration	0.345
Fibrinogen and Hypertension	0.290
Fibrinogen and duration of Diabetes	0.392
Hypertension and duration of Diabetes	0.341
Fibrinogen and Hypertension and duration of Diabetes	0.394
Fibrinogen, Hypertension, duration of Diabetes and HBA_1C	0.389
Fibrinogen, Hypertension, duration of Diabetes , HBA_1C and cholesterol All	0.387

Other parameters entered in analyses included cholesterol, triglycerides, HDL &LDL cholesterol, age, sex, glucose, type of treatment, smoking, and hypertension.

Fibrinogen and duration were individually significant determinants (r2 = 0.289 and 0.345, respectively; P < 0.002). hypertension by itself, was a relatively minor determinant (r2 = 0.040); however, addition of hypertension to duration and fibrinogen raised their predictive power to ~39% (P< 0.001).

DISCUSSION

The search for the factor(s) underlying accelerated complication in diabetes continues. It is clear that traditional risk factors for atherosclerosis one of macrovascular complication e.g., age, hyperlipidemia, smoking, hypertension, and obesity do not explain this accelerated risk (11, 12). Other potential candidates include platelet hypersensitivity (24), coagulation factors (25), and perhaps endothelial cellular dysfunction (26). The incriminatory evidence for these additional mechanisms is, as yet, far from conclusive.

In this study, we attempted to explore further the prevalence and role of fibrinogen in relation to diabetic vascular disease, in view of the existing epidemiological evidence in general populations (1-4). A role for fibrinogen in the pathophysiology of diabetes was suggested by the reports of 1) hyperfibrinogenemia in cross-sectional studies, albeit in a small number of patients (14- 18); 2) impaired fibrinolysis at rest and/or after physical training in several, but not all, studies (8,18,25,27); 3) enhanced generation of fibrin degradation products (28,29); 4) inhibition of antithrombin III activity by nonenzymatic glycosylation, although in the setting of unphysiological glucose concentrations (30); and 5) reduced fibrinogen survival (28,31). The results presented in this report indicate that fibrinogen concentrations frequently are elevated in diabetes, and type II and duration, but particularly

in those with type I diabetes and preexisting vascular complications. However, even patients with type I diabetes and no clinically evident vascular complications had significantly elevated mean fibrinogen levels. Evidence for elevated fibrinogen concentrations before the onset of vascular disease is controversial, primarily because of difficulties in clinically assessing onset. In a small number of patients with > 15 yr duration of type I diabetes, Coller et al. (16) found no appreciable increase in fibrinogen levels in those without retinopathy (n = 8), whereas those with retinopathy (n = 21) had striking elevations. Similarly, Jensen et al. (32) reported a progressive increase in fibrinogen levels with increasing severity of proteinuria compared with those with no proteinuria. On the other hand, fibrin degradation products were elevated markedly in newly diagnosed type 1 patients in another report (29), and elevated fibrinogen levels were documented even in mildly glucose intolerant diabetic patients by McMillan et al. (17). Taken together, these observations and ours support the view that fibrinogen excess might contribute not only to the vessel wall pathology (5,6), but it may even exacerbate the diabetic vasculopathy, rather than simply being a marker for preexisting disease .

Another important issue is the relationship of fibrinogen levels with hyperglycemia. We found significant correlations between fibrinogen and ambient plasma glucose levels and these index of diabetes control have discernible impact on the prediction of fibrinogen in multiple regression model (Table 5). However, note that Jones et al. (28,31) found that shortened fibrinogen survival was reversible by correction of hyperglycemia. Therefore, it is possible that hyperglycemia play an important role in thrombin activation in the presence of elevated fibrinogen concentrations.

Of considerable interest are the findings in our study of the relationship of fibrinogen with vascular complications (Tables 3 and 6). Not surprisingly, the patients with

complications (group B) were older, had longer duration of diabetes, poorer control, and higher cholesterol levels. The mean fibrinogen level in this group was 47% greater compared with control subjects. In view of the interdependence of multiple predictors of vascular complications, the stepwise correlations in multiple regression analyses (Table 6) are of particular interest. As indicated by these analyses, vascular complications correlated best with only 3 of 14 factors entered in this model, i.e., fibrinogen, hypertension, and duration of diabetes.

Individually, fibrinogen appears to be as important as duration and hypertension, and together, these three variables account for ~ 39% of the predictability of the presence of vascular complications.

CONCLUSION

In summary, our study suggest that change fibrinogen level may be one of the important missing links in the pathogenesis of diabetic vascular disease. Hyperfibrinogenemia is particularly prevalent in type II diabetes, and along with hypertension and duration of diabetes, it may be a major independent predictor of vascular complications. Future studies of pathogenesis and prevention of diabetic vascular disease should be designed to evaluate the role of fibrinogen, among other known and potential risk factors.

ACKNOWLEDGEMENT:

Special thanks to the Staff of Haematology Department, Faculty of Medical Laboratory Sciences, Alneelain University, and we appreciate also Ahmed S. Kh. Adam who contribute in manuscript preparation. Finally special thanks to the patients for being cooperative, despite their pain.

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