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Full Length Research Article

Evaluation of Prothrombin Time and Activated Partial Thromboplastin in Patients with Diabetes Mellitus

[*1]Y. Abdulrahaman and [2]M.K. Dallatu
[1] Haematology Department, School of Medical Laboratory Science, Usmanu Danfodiyo University Sokoto.
[2]Chemical Pathology Department, School of Medical Laboratory Science, Usmanu Danfodiyo University Sokoto
[*Author of correspondence yakubujos2006@yahoo.com; ☎: +2348034736809]

ABSTRACTS: Diabetes mellitus is a heterogeneous disorder that affects cellular metabolism in variety of ways, and coagulation indices are reported to be adversely affected. In the current study prothrombin time (PT) and activated partial thromboplastin time (APTT) were investigated in treated and untreated diabetics as well as in non diabetic controls. Using clot based assay technique, PT was 20.620± 2.849, 16.720±2.339, and 14. 920±1.209 in untreated, treated and in non diabetic controls respectively. There were statistically significant differences ($p<0.05$) between the untreated and treated diabetics as well as between untreated and non diabetic controls. APTT was 58.460±4.146, 43.260±5.587 and 41.380±4.295 in untreated, treated and in non diabetic controls respectively. There is statistically significant difference ($p>0.05$) between untreated and treated diabetics ($p<0.05$).Value of treated and controls are not significantly difference ($p>0.05$). Our finding suggest an abnormal PT and APTT in untreated patients with diabetes mellitus.
Keywords: Diabetes, Prothrombine Time, Activated Partial Thromboplastin Time

INTRODUCTION

Diabetes mellitus is a common endocrine disease of multiple aetiology (Oguntona and Amballi, 2005; Ogunkolo and Alebiosu, 2006). It is characterised by chronic hyperglycaemia with subsiquent disturbances of carbohydrates, fat and protein metabolism (Momo *et al.,* 2006). Excessive blood glucose produces the classical symptoms such as polydipsia (increased thirst), polyuria (frequent urination) and polyphagia (increased hunger) (Cooke and Plotnick , 2008). In Nigeria and the world at large, Diabetes is a major health problem with about 90% of diabetic patients having non insulin type II while about 10% have insulin dependent (Ohwworiola *et al*., 1998).

Body of evidence suggest that certain haematological indices are altered in patients with diabetes mellitus (Dallatu *et al*., 2010). In patient with diabetes mellitus, persistent hyperglycaemia exposes red blood cells (RBCs) to elevated glucose concentration, thus resulting in glycalation of haemoglobin, prothrombin, fibrinogen and other proteins involved in clotting mechanisms (Selvin *et al*., 2010). The glycation results in the incomplete activation and function of the clotting cascade (Qin, *et al*., 2004). Glycation of intrinsic and extrinsic clotting proteins will decrease the availability of these proteins which affect the clotting capacity (Lippi *et al*., 2009).

Prothrombin time (PT) and activated partial thromboplastin time (APTT) are haematological indices that give an insight into the coagulation status of patients (Hinchcliff *et al.,* 2004). Divided into 3 pathways of intrinsic, extrinsic and common, these factors collectively play a central role in arresting bleeding disorder (Merlo *et al*., 2002, Kucharska-Newton *et al*., 2009).

The blood contains about a dozen clotting factors (Alesci *et al*., 2008). These factors are proteins that exist in the blood in an inactive state, but can be called into action when tissues or blood vessels are damaged (Alesci *et al*., 2008). Blood clotting is the transformation of liquid blood into a semisolid gel. Clots are made from fibers (polymers) of a protein called fibrin. Fibrin monomers come from an inactive precursor called fibrinogen. Also called Factor I, fibrinogen play critical role in blood viscosity. Increased concentration of fibrinogen (hyperfibrinogenemia) in uncontrolled NIDDM patients is implicated in vascular damage induction (Zachary and Bloomgarden, 2011).

In the laboratory, measurement of PT, APTT, and fibrinogen concentration are the most commonly employed laboratory tests in patients with a suspected coagulopathy (Furlanello *et al*., 2006). Prothrombin time is a laboratory screening test used to detect disorders involving the activity of the factors I, II, V, VII, and X of the extrinsic and common pathways (Hinchcliff *et al.,* 2004). Activated partial thromboplastin time is used to screen for abnormalities of the intrinsic and common clotting systems and to monitor the anticoagulant effect of circulating heparin. It measures the activities of factors I, II, V, VIII, IX–XI, and XII of the intrinsic and common pathways (Iazbik *et al.*, 2001).

Changes in these proteins favour the development of hyper-coagulable and pro-thrombotic state, which may in turn enhance cardiovascular risk by increasing the likely hood of developing an occlusive thrombus within a coronary/cerebral artery contributing to the development of atherosclerotic lesion (Dunn and Grant, 2005). PT and APTT can therefore be used to assess the risk of clotting complications in patients with diabetes mellitus although modern coagulation diagnostic test are becoming more sophisticated, PT and APTT are still important basic examinations in clinical laboratories (Ng, 2009).

The current study was therefore designed to evaluate the PT and APTT in treated, untreated and in non diabetics control subjects.

MATERIALS AND METHODS

One hundred and fifty (150) diabetic patients of 50 treated, 50 untreated and 50 apparently healthy non diabetic controls where selected for the research. They were recruited from patients attending diabetes clinic of Usmanu Danfodiyo University Teaching Hospital, Sokoto, North West Nigeria. Institutional ethical clearances were obtained and an informed consent form was explained to each participant before they signed the consent. Specimens for PT and APTT measurement were obtained by vein puncture after a 12 hour fast. PT and APTT were measured using calcium rabbit brain thromboplastin and kaolin platelet substitute techniques (DIAGEN DIAGNOSTIC REAGENT LTD, OXON, U.K). Briefly, PT was assayed with two hundred microlitre of calcum rabbit brain thromboplastin reagent placed in a clotting tube and incubated in a water bath at 37^{o} C for 2 minutes. Hundred microlitre of plasma is then added and a stop watch started. The tube is gently tilted at regular interval and the watch was stopped when the clot formation was observed. For APTT, two hundred microlitre of kaolin platelet substitute mixture was placed in a clotting tube and incubated in a water bath at 37^{o}C for 2 minutes. One hundred microliter of plasma was then added and the tube was gently tilted at interval for exactly two minutes. One hundred microliter of calcium chloride (pre-incubated at 37^{O} C) was then added and a stop watch started. The tube was tilted at intervals and the time for clot formation was recorded. Our laboratory has a reference ranges for PT and APTT, as 10-16 seconds and 30-48 seconds respectively.

Statistical Analysis

Data processing was done using one way ANOVA Graphic Pad Instant software package, Sandiego, U.S.A. The result was expressed as mean ± SD. The paired t-test was used to determine the significant level. $p<0.05$ was considered as statistically significant.

RESULTS

Result of prothrombin time (PT) and activated partial thromboplastin time (APTT) study was shown in Table 1. For PT, there are statistically significant differences between the untreated and treated diabetics as well as between untreated and non diabetic controls ($p<0.05$). The difference between treated and non diabetics controls is not significant. For APTT, there is statistically significant difference ($P<0.05$) between untreated and treated diabetics. The difference between treated and non diabetic controls is not statistically significant ($p>0.05$).

Table 1: Prothrombin Time (PT) and Activated Partial Thromboplastin Time (APTT) concentrations of study subjects in Sokoto, Nigeria

GROUPS	PT(seconds)	APTT(seconds)
Untreated diabetic	20.620±2.849*	58.460±4.146*
Treated diabetics	16.720±2.339,	43.260±5.587
Control	14.920±1.209	41.380±4.295

Value are Mean ± SD *significantly different ($P<0.05$) when compared with non diabetic controls using Graphic Pad Instant software, Sandiego, U.S.A

DISCUSSION

Diabetes mellitus is a heterogeneous disease affecting metabolism of various compounds including carbohydrates, lipids and proteins which also impairs biological processes such as coagulation homeostasis that causes vascular thrombotic problems (Carr, 2001; Hameed *et al*., 2002).

The circulatory disturbances in diabetes are characterized by alternation in platelet count and activity, coagulopathy, fibrinolytic aberration, haemorrheologic factors and changes in endothelial metabolism (McFarlane, 1997). In patient with diabetes mellitus, persistent hyperglycaemia exposes red blood cells (RBC) to elevated glucose concentration, thus resulting in glycation of haemoglobin, prothrombin, fibrinogen and other protein involved in clotting mechanisms (Selvin *et al*., 2010).

In the current study, it was observed that non-diabetic individuals and treated diabetics have a prothrombin time (PT) value within the reference range of 10-16 as against a value of 20.620±2.849 seen in untreated diabetics. This is in agreement with the findings of (Alao *et al*., 2010). In patients with diabetes mellitus,

abnormalities in coagulation haemostasis, platelets dysfunction and reduced activity of fibrinolytic system can collectively accelerate atherogenesis in diabetic patients (Carr, 2001). Merlo *et al*., (2002) reported an increase in tissue factor (TF) and subsequent conversion of inactive factor vii to active factor vii which triggers the extrinsic pathway . This will lead to an increase in PT as reported here. Diabetic hypertriglyceridemia, the additional risk factor for cardiovascular problems, in addition to increased levels of TF and an activated factor VII gives a bad prognosis for serious fatal ischemic heart disease (Kucharska-Newton *et al*., 2009). Hyperglycemia has been considered to be the causative factor of these abnormalities in coagulation pathways, and exposes patient with diabetes to macrovascular mortality (Standl *et al*., 1996). Several studies have shown that levels of PT are increased in both type 1 and type 2 diabetic patients and that glycaemic control normalizes PT levels in diabetic patients (Standl *et al*, 1996).

In the current study, a significant elevation of APTT was recorded in untreated diabetics when compared with treated diabetics ($p<0.05$). This is similar to the findings of Hassan, (2009) and Maysam *et al*., (2011). Increased plasma levels of PT and APTT are consistent with abnormal coagulation mechanism and may be interpreted as a tendency to bleeding and cardiovascular disorders (Hassan, 2009). Increase in the intrinsic pathway proteins, and activation of blood coagulation mechanism are consistent in diabetics (Berliner *et al*., 2002; Eteng *et al*., 2008; Behnam *et al*., 2010).

The pathogenetic mechanism of the clotting activation in diabetes is not completely clear. Perturbance of components of the anticoagulant system associated with hyperglycaemia may play an important role as exemplified in hyperglycaemia induced depression of biological activity of the anticoagulant proteins such as AT-III. This, in addition to non enzymatic glycation, are suggested to be the causative factors of AT-III dysfunction (Hassan, 2009).

The presents finding and previous studies (Hu, *et al*., 1998). Thrombotic and hypercoagulable complications in diabetes patients are introductions to vascular and cardiovascular complications (Gabazza *et al*., 1996). A prognostic clue in the simultaneous measurement of factors I, II, V, VII, X for PT and factors I, II, V, VII, X, Phospholipids and calcium for APTT and fibronogen could be initially prescribed. Since these factors show critical and more specific alterations in the early stages of diabetes disease, by giving an awfully bad prognosis in coagulation disturbance. Therefore, hypercoagulable state management may have a preventive value in subsequent vascular complications in patients with diabetes mellitus

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