DETERMINATION OF PROTEIN C LEVEL IN TYPE 2 DIABETIC PATIENTS IN SUDAN

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ABSTRACT
Protein C is a precursor to a serine protease present in plasma that plays an important physiological role in the regulation of blood coagulation. Protein C deficiency causes thrombophilia or hypercoagulability and increased risk of venous thrombosis. The aim of this study was to measure protein C level among type 2 diabetes mellitus patients in Sudan. A total of 90 samples were collected (45 patients with type 2 DM and 45 healthy controls), 50 were males and 40 were females, their age ranged between 18 to 52 years. The protein C level was measured by Enzyme Linked Immunosorbent Assay ELIZA method, to correlate the protein C level with patient’s age, gender and HbA1c. Data were analyzed by using statistical package for the social science (SPSS). The present study showed that mean PC level among case group (50±12.4) was significantly lower than control group (83±12 (P-value 0.000).

KEYWORDS: Protein C, Type 2 diabetic mellitus, Sudan.

INTRODUCTION
Diabetes mellitus (DM) is characterized by hyperglycemia accompanied with the biochemical alterations in carbohydrate, protein and lipid metabolism.[1] Due to the increasing number of diabetics, this disease has acquired a character of “epidemic” in recent decades. In 2000, the number of diabetics worldwide was approximately 151 million, estimates are that in 2010 this will reach 221 million and by 2025, 324 million.[2] It is believed that changes in human behavior, environment and lifestyle are favoring an increase in the number of obese and diabetic individuals.[3] According to WHO, diagnostic criteria of diabetes mellitus are fasting blood glucose ≥7.0 mmol/l, and 2 hours after glucose <1.11mmol/l and HbA1c <6.5%.[4] There are Two types of diabetes mellitus are the most prevalent: type-1 diabetes is characterized by autoimmune destruction of pancreatic beta cells resulting in an absolute deficiency in insulin, and type 2 diabetes (T2DM or adult-onset), which corresponds to approximately 90% of cases of diabetes worldwide, is characterized by insulin resistance and/or reduced production of insulin.[5] Patients suffering from diabetes mellitus a high probability of developing acute cardiovascular disease, in particular myocardial infarct and cerebrovascular stork.[6] Cardiovascular disease (CVD) is the leading cause of disability and premature mortality in patients with diabetes.[7] About 80% patients with diabetes may die due to thromboembolic CVD.[8] Much attention has been devoted to the pathogenic factors, altered haemostatic balance, including abnormalities in platelet function, increase in blood coagulability and altered fibrinolytic system.[9] Since most of these abnormalities indicate a hypercoagulable and hypofibrinolytic state leading to enhance intravascular fibrin deposition they have been linked to the high risk for cardiovascular morbidity and mortality among these patients.[10] Coagulation abnormalities with decreased level of antithrombin III, protein C and protein S has been reported in DM with elevated clotting factors.[11] Protein C is a precursor to a serine protease present in plasma that plays an important physiological role in the regulation of blood coagulation.[12] Protein C is a vitamin K dependent factor, which, in its active form, inactivates coagulation factor Va and VIIIa. Protein S, another vitamin K dependent factor serves as a cofactor of activated protein C.[13] APC was demonstrated to show profibrinolytic activity by inactivating plasminogen activator inhibitor. Since severe thrombosis was observed in patients with protein C deficiency. Protein C might play a critical role in antithrombotic process. Protein C might be consumed by developing the hypercoagulable state. Two types of PC deficiency states...
are recognized, in type I deficiency the plasma concentration of PC is reduced both in functional and immunological assay, reflecting a genetic defect causing a reduced biosynthesis of PC. Type II deficiency is characterized by normal protein C antigen levels, but with decreased functional activity. This type of defect reflect the synthesis of abnormal molecules with reduced function, the mutation in the PC gene have been characterized in a recently published data base.\textsuperscript{[14]} Because of the critical role that protein C play as an anticoagulant those deficiencies in PC or APC resistance lead to significant risk of forming blood clots.

MATERIALS AND METHODS
This was a case-control study, conducted in Khartoum, Sudan during the period of April 2017 to August 2017. 90 samples were included (45 samples of type 2 diabetic patient's individuals and 45 samples of healthy individuals as control).

Five ml of venous blood was collected: 2.5 ml in 3.8% Trisodium citrate (1:9 volume of sodium citrate solution), and 2.5 in EDTA container. Haemoglobin A1c (HBA1c) was measured using NYco Card READER II.

Protein C was measured using Enzyme Linked Immunosorbent Assay ELISA commercial assay kit from Aesku, Diagnostics, Germany (normal range: 70-140%). Data were analyzed by using statistical package for the social science (SPSS) Version 23.

RESULTS
This study was conducted in Khartoum Sudan Among 90 individuals. There were divided into two groups 45 were patients with type 2 diabetes mellitus who considered as cases and 45 apparently healthy individuals who considered as control. Our study revealed statistically significant difference between cases and controls concerning protein C concentration. Mean PC level among case group (50±12.4) was significantly lower than control group (83±12 (P-value 0.000), as shown in table 4.1. While there was no correlation between Protein C concentration and HbA1c Concentration among study group (R value=0.145, P value=0.349) as shown in figure 4.1.

Table (4.1): Protein C Level among study population (CASE & control).

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Case NO (45)</th>
<th>Control NO (45)</th>
<th>P. Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Protein C Mean (SD)</td>
<td>50 ± (12.4)</td>
<td>83± (12)</td>
<td>0.000</td>
</tr>
</tbody>
</table>

Ninety patients were enrolled in this study (45 were diabetic patients and the others were healthy controls). Our study revealed statistically significant difference between cases and controls concerning protein C level (Mean=50 ± 12.4 and 83± 12, P value=0.000 respectively). While there was no correlation between Protein C and HbA1c concentration among study group (R value=0.145, P value=0.349). Also non-significant association between protein C level and age and gender was detected (p value= 0.739 and gender p value=0.325).

In our study the mean of protein C level was significantly lower than that of the healthy control group (P < 0.00) Our observation was matched with berne ASLAN\textsuperscript{[15]} and Nantarat komanasin\textsuperscript{[16]}, Ceriello,\textsuperscript{[17]} Saito et al.,\textsuperscript{[18]} found that the PC-Ag levels of the diabetic patients were higher than that of the control group.

These finding suggested that increased thrombin production in diabetes mellitus causes enhanced activation of protein C in the circulation and in turn enhanced clearance from the blood.\textsuperscript{[17]}

This study revealed low protein C in type 2 diabetic patients, the decrease in protein C level is due to improper synthesis of protein C in the liver. Also decrease in protein C level maybe due to the increase in clearance in diabetic patients. This abnormal decrease in protein C level promote the hypercoagubale state in diabetic patients which leads to venous thrombosis.

DISCUSSION
Diabetes mellitus patients have a high probability of developing acute cardiovascular disease, in particular myocardial infarction and cerebrovascular stroke.

It has been suggested that diabetes can be associated with hypercoagulability in abroad sense, which is related to the development of vascular complication.
CONCLUSION
The study findings concluded that plasma protein C level decreases in patients with diabetic mellitus and these changes are not influenced HbA1c.

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REFERENCES
15. Berna ASLAN, Nezaket EREN, Pbnem CÜÜERLÜ, Fatma M.I.D.R, Nihal Y.CELResearch and Training Hospital, Biochemistry Laboratory, Üstanbul – TurkeyReceived, 2004 April 15.