International Journal of Multidisciplinary and Current Research ISSN: 2321-3124
Research Article Available at: http://ijmcr.com

Assessment of Coagulation Process in Diabetic Patients using Prothrombin Time and Activated Thromboplastin Time Tests

Abdulla Musa Abdulla[1*], Tariq.E.Elmissbah[2], Esra Mohammed Hamid[3], Fatima Osman A-ltom[3] and Maisa Faisal Abusham[3]

[1]Department of Medical Laboratory Sciences, College of Applied Medical Sciences, Shaqra University, KSA
[2]Department of Medical Laboratory Sciences, College of Applied Medical Sciences, Taif University, KSA
[3]College of Medical Laboratory Sciences, Sudan University of Sciences and Technology Khartoum- Sudan

Accepted 01 April 2017, Available online 07 April 2017, **Vol.5 (March/April 2017 issue)**

Abstract

Background: *Diabetes mellitus (DM), commonly referred to as diabetes, is a group of metabolic diseases in which there are high blood sugar levels over a prolonged period.*
Study design: *This study was designed to determine effects of diabetes on coagulation process using prothrombin time test (PTT), and activated partial thromboblasin time test (APTT) and consent was taken from the health authority.*
Materials and methods*: Hundred blood samples were collected into EDTA blood containers 5 ml of venous blood were collected in plastic container containing 2.8 ml of aqueous tri-sodium citrate as anticoagulant. Blood was mixed and centrifuged at 4000 rpm for ten minutes, and platelet poor plasma was collected. PT and APTT were estimated using coagulometer.*
Results*: The results showed the mean of PT in diabetes was 12.609 in patients when was 13.905 in control, when the mean of APTT was32.745 in diabetic patients, when was 32.745 in control*
Conclusion: *From the present study it may concluded that diabetes mellitus had no effects on PT and APTT.*

Keywords*: Diabetes Mellitus, Prothrombin Time, Activated Partial Thromboblastin Time.*

Introduction

Diabetes mellitus (**DM**), commonly referred to as **diabetes**, is a group of metabolic diseases in which there are high blood sugar levels over a prolonged period.[2] Symptoms of high blood sugar include frequent urination, increased thirst, and increased hunger. If left untreated, diabetes can cause many complications.[3] Acute complications include diabetic ketoacidosis and nonketotic hyperosmolar coma.[4] Serious long-term complications include cardiovascular disease, stroke, chronic kidney failure, foot ulcers, and damage to the eyes.[3]

Diabetes is due to either the pancreas not producing enough insulin or the cells of the body not responding properly to the insulin produced.[5] There are three main types of diabetes mellitus:

- Type 1 DM results from the pancreas' failure to produce enough insulin. This form was previously referred to as "insulin-dependent diabetes mellitus" (IDDM) or "juvenile diabetes". The cause is unknown.[3]
- Type 2 DM begins with insulin resistance, a condition in which cells fail to respond to insulin properly.[3] As the disease progresses a lack of insulin may also develop.[6] This form was previously referred to as "non insulin-dependent diabetes mellitus" (NIDDM) or "adult-onset diabetes". The primary cause is excessive body weight and not enough exercise.[3]
- Gestational diabetes, is the third main form and occurs when pregnant women without a previous history of diabetes develop a high blood sugar level.[3]

Type 1

Type 1 diabetes mellitus is characterized by loss of the insulin-producing beta cells of the islets of Langerhans in the pancreas, leading to insulin deficiency. This type can be further classified as immune-mediated or idiopathic. The majority of type 1 diabetes is of the immune-mediated nature, in which a T-cell-mediated autoimmune attack leads to the loss of beta cells and thus insulin.[26] It causes approximately 10% of diabetes mellitus cases in North America and Europe. Most affected people are otherwise healthy and of a healthy weight when onset occurs. Sensitivity and responsiveness to insulin are usually normal, especially in the early stages. Type 1 diabetes can affect children or adults, but was

Corresponding author's ORCID ID: 0000-0000-0000-0000
DOI: https://doi.org/10.14741/ijmcr/v.5.2.14

Type 2

Type 2 diabetes mellitus is characterized by insulin resistance, which may be combined with relatively reduced insulin secretion.[5] The defective responsiveness of body tissues to insulin is believed to involve the insulin receptor. However, the specific defects are not known. Diabetes mellitus cases due to a known defect are classified separately. Type 2 diabetes is the most common type.

In the early stage of type 2, the predominant abnormality is reduced insulin sensitivity. At this stage, hyperglycemia can be reversed by a variety of measures and medications that improve insulin sensitivity or reduce glucose production by the liver.

Type 2 diabetes is due primarily to lifestyle factors and genetics.[29] A number of lifestyle factors are known to be important to the development of type 2 diabetes, including obesity (defined by a body mass index of greater than thirty), lack of physical activity, poor diet, stress, and urbanization.[12] Excess body fat is associated with 30% of cases in those of Chinese and Japanese descent, 60–80% of cases in those of European and African descent, and 100% of Pima Indians and Pacific Islanders.[5] Even those who are not obese often have a high waist–hip ratio.[5]

Gestational diabetes

Gestational diabetes mellitus (GDM) resembles type 2 diabetes in several respects, involving a combination of relatively inadequate insulin secretion and responsiveness. It occurs in about 2–10% of all pregnancies and may improve or disappear after delivery.[34] However, after pregnancy approximately 5–10% of women with gestational diabetes are found to have diabetes mellitus, most commonly type 2.[34] Gestational diabetes is fully treatable, but requires careful medical supervision throughout the pregnancy. Management may include dietary changes, blood glucose monitoring, and in some cases insulin may be required.

Pathophysiology

Insulin is the principal hormone that regulates the uptake of glucose from the blood into most cells of the body, especially liver, muscle, and adipose tissue. Therefore, deficiency of insulin or the insensitivity of its receptors plays a central role in all forms of diabetes mellitus.[38]

The body obtains glucose from three main places: the intestinal absorption of food, the breakdown of glycogen, the storage form of glucose found in the liver, and gluconeogenesis, the generation of glucose from non-carbohydrate substrates in the body.[39] Insulin plays a critical role in balancing glucose levels in the body. Insulin can inhibit the breakdown of glycogen or the process of gluconeogenesis, it can stimulate the transport of glucose into fat and muscle cells, and it can stimulate the storage of glucose in the form of glycogen.[39]

Insulin is released into the blood by beta cells (β-cells), found in the islets of Langerhans in the pancreas, in response to rising levels of blood glucose, typically after eating. Insulin is used by about two-thirds of the body's cells to absorb glucose from the blood for use as fuel, for conversion to other needed molecules, or for storage. Lower glucose levels result in decreased insulin release from the beta cells and in the breakdown of glycogen to glucose. This process is mainly controlled by the hormone glucagon, which acts in the opposite manner to insulin.[40]

Hemostasis is a process which causes bleeding to stop, meaning to keep blood within a damaged blood vessel (the opposite of hemostasis is hemorrhage). It is the first stage of wound healing. This involves coagulation, blood changing from a liquid to a gel. Intact blood vessels are central to moderating blood's tendency to form clots. The endothelial cells of intact vessels prevent blood clotting with a heparin-like molecule and thrombomodulin and prevent platelet aggregation with nitric oxide and prostacyclin. When endothelial injury occurs, the endothelial cells stop secretion of coagulation and aggregation inhibitors and instead secrete von Willebrand factor which initiate the maintenance of hemostasis after injury. Hemostasis has three major steps: 1) vasoconstriction, 2) temporary blockage of a break by a platelet plug, and 3) blood coagulation, or formation of a fibrin clot. These processes seal the hole until tissues are repaired.

Hemostasis occurs when blood is present outside of the body or blood vessels. It is the instinctive response for the body to stop bleeding and loss of blood. During hemostasis three steps occur in a rapid sequence. Vascular spasm is the first response as the blood vessels constrict to allow less blood to be lost. In the second step, platelet plug formation, platelets stick together to form a temporary seal to cover the break in the vessel wall. The third and last step is called coagulation or blood clotting. Coagulation reinforces the platelet plug with fibrin threads that act as a “molecular glue”.[1] Platelets are a large factor in the hemostatic process. They allow for the creation of the “platelet plug” that forms almost directly after a blood vessel has been ruptured. Within seconds of a blood vessel’s epithelial wall being disrupted platelets begin to adhere to the sub-endothelium surface. It takes approximately sixty seconds until the first fibrin strands begin to intersperse among the wound. After several minutes the platelet plug is completely formed by fibrin.[2] Hemostasis is maintained in the body via three mechanisms:

1. **Vascular spasm** - Damaged blood vessels constrict. Vascular spasm is the blood vessels' first response to injury. The damaged vessels will constrict (vasoconstrict) which reduces the amount of blood flow through the area and limits the amount of blood loss. This response is triggered by factors such as a direct injury to vascular smooth muscle, chemicals released by endothelial cells and platelets, and reflexes initiated by local pain

receptors. The spasm response becomes more effective as the amount of damage is increased. Vascular spasm is much more effective in smaller blood vessels.[1]

2. **Platelet plug formation** - Platelets adhere to damaged endothelium to form a platelet plug (*primary hemostasis*) and then degranulate. This process is regulated through thromboregulation. Plug formation is activated by a glycoprotein called Von Willebrand factor (vWF), which is found in plasma. Platelets play one of the biggest roles in the hemostatic process. As they adhere to the collagen fibers of a wound, platelets become spiked and much stickier. They then release chemical messengers such as adenosine diphosphate (ADP), serotonin and thromboxane A2, causing more platelets to stick to the area, release their contents, and enhance vascular spasms. As more chemicals are released more platelets stick and release their chemicals; creating a platelet plug and continuing the process in a positive feedback loop. Platelets alone are responsible for stopping the bleeding of unnoticed wear and tear of our skin on a daily basis.[3][4][5]

There are a dozen proteins that travel along the blood plasma in an inactive state and are known as clotting factors. Once the platelet plug has been formed by the platelets, the clotting factors begin creating the clot. When this occurs the clotting factors begin to form a collagen fiber called fibrin. Fibrin mesh is then produced all around the platelet plug, which helps hold the plug in place. Once this begins, red and white blood cells become caught up in the fibrin mesh which causes the clot to become even stronger.[3]

3. **Blood coagulation** - Clots form upon the conversion of fibrinogen to fibrin, and its addition to the platelet plug (*secondary hemostasis*). Coagulation: The third and final step in this rapid response reinforces the platelet plug. Coagulation or blood clotting uses fibrin threads that act as a glue for the sticky platelets. As the fibrin mesh begins to form the blood is also transformed from a liquid to a gel like substance through involvement of clotting factors and pro-coagulants. The coagulation process is useful in closing up and maintaining the platelet plug on larger wounds. The release of prothrombin also plays an essential part in the coagulation process because it allows for the formation of a thrombus, or clot, to form. This final step forces blood cells and platelets to stay trapped in the wounded area. Though this is often a good step for wound healing, it has the ability to cause severe health problems if the thrombus becomes detached from the vessel wall and travels through the circulatory system; If it reaches the brain, heart or lungs it could lead to stroke, heart attack, or pulmonary embolism respectively. However, without this process the healing of a wound would not be possible.[1]

Materials and Methods

Subjects

In this study eighty samples were collected from diabetic patients and twenty samples from healthy participants as control.

Sample

2.5 ml of venous blood were collected in plastic container containing 2.8 ml of aqueous tri-sodium citrste as anticoagulant. Blood was mixed and centrifuged at 4000 rpm for ten minutes, and platelet poor plasma was collected.

Principle of prothrombin time (PT), and activated thromoblastin time (APTT)

The PT measyres the clotting time of plasma in the presence of an optimal concentration of tissue extract (thromboplastin) and indicates the overall efficiency of the extrinsic coagulation system [4]

Results

Table.1 Percentage of case and control among study

	%
Case	**80**
Control	**20**

Table 2 Mean of PT among case and control

	PT		
	N	**Mean**	**P.Value**
Case	80	12.609	.004
Control	20	13.905	

Table 3 Mean of APTT among case and control

	APTT		
	N	**Mean**	**P.Value**
Case	80	32.745	.000
Control	20	32.745	

Table 4 Mean of PT among sex, treatment, age group and duration group in cases

	PT			
		N	**Mean**	**P.Value**
Sex	Male	32	13.031	.191
	Female	48	12.327	
Treatment	Yes	72	12.490	.178
	No	8	13.675	
Age group	≥ 40	46	12.598	.962
	≤ 40	34	12.624	
Duration group	≥ 10	16	13.131	.323
	≤ 10	64	12.478	

Table 5 Mean of PT among sex, treatment, age group and duration group in cases

		APTT		
		N	**Mean**	**P.Value**
Sex	Male	32	33.622	.905
	Female	48	32.160	
Treatment	Yes	72	32.718	.267
	No	8	32.988	
Age group	≥ 40	46	32.098	.267
	≤ 40	34	33.621	
Duration group	≥ 10	16	32.012	.410
	≤ 10	64	32.928	

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 thromboticproblems (Carr, 2001; Hameed *et al*., 2002). Patients with diabetes mellitus persistent hyperglycemia exposes red blood cells to elevated glucose concentration, resultion in glycation of hemoglobin, prothrombin fibrinogen, and other protein involved in clotting mechanism

The present study was conducted to evaluate and compare coagulation tests among T2DMand healthy individuals. DM is characterized by high risk of atherothrombotic complications affecting the coronary, cerebral and peripheral arterial trees. It is a syndrome characterized by presence of chronic hyperglycemia due to defective insulin secretion, insulin action or both affecting metabolism of various compounds including carbohydrate, lipids, and proteins and it also impairs various biological processes such as coagulation and fibrinolytic alteration. In this study PT had a significant variation among case and control (P.value.0.004,less than0.05).Also PT was insignificant in patients of all duration of disease.(P.value 0.323,which more than 0.05).Also in this study PT was insignificant in patients with different ages. PT had insignificant variation with in diabetic patients with and without treatment. The level of APTT level in this study showed significance variation among case and control ((P.value.0.000,less than00.05), but within normal ranges, Also study proved that APTT showed insignificant variation according to the duration of disease, and also APTT showed insignificant according to the ages , and treatment of patients. These finding strongly agreed with many studies similar to this study.[17, 18, 19]

Conclusion

From the present study it may concluded that diabetes mellitus had no effects on PT and APTT.

Conflict of Interest: None.

References

[1]. Clement S, Braithwaite SS, Magee MF, Ahmann A, Smith EP, *et al*. American Diabetes Association Diabetes in Hospitals Writing Committee Management of diabetes and hyperglycemia in hospitals. Diabetes Care. 2004;27:553–591. [PubMed]

[2]. Lemkes BA, Hermanides J, Devries JH, Holleman F, Meijers JC, *et al.* Hyperglycaemia, a prothrombotic factor? J Thromb Haemost. 2010;8:1663–1669. [PubMed]

[3]. Bick RL, Arun B, Frenkel EP. Disseminated intravascular coagulation. clinical and pathophysiological mechanisms and manifestations. Haemostasis. 1999;29:111–134. [PubMed]

[4]. Ng VL. Prothrombin time and partial thromboplastin time assay considerations. Clin Lab Med. 2009;29:253–263. [PubMed]

[5]. Tripodi A, Chantarangkul V, Martinelli I, Bucciarelli P, Mannucci PM. A shortened activated partial thromboplastin time is associated with the risk of venous thromboembolism. Blood. 2004;104:3631–3634.[PubMed]

[6]. Lippi G, Franchini M, Targher G, Montagnana M, Salvagno GL, *et al.* Epidemiological association between fasting plasma glucose and shortened APTT. Clin Biochem. 2009;42:118–120. [PubMed

[7]. Grant PJ. Diabetes mellitus as a prothrombotic condition. J Intern Med. 2007;262:157–172. [PubMed]

[8]. Reddy NM, Hall SW, MacKintosh FR. Partial thromboplastin time: prediction of adverse events and poor prognosis by low abnormal values. Arch Intern Med. 1999;159:2706–2710. [PubMed]

[9]. Anand SS, Yi Q, Gerstein H, Lonn E, Jacobs R, *et al*. Study of Health Assessment and Risk in Ethnic Groups; Study of Health Assessment and Risk Evaluation in Aboriginal Peoples Investigators Relationship of metabolic syndrome and fibrinolytic dysfunction to cardiovascular disease. Circulation. 2003; 108:420–425. [PubMed]

[10]. American Diabetes Association. Standards of medical care in diabetes-2010. Diabetes Care. 2010;33:S11–61. [PMC free article] [PubMed]

[11]. Alberti KG, Zimmet PZ. WHO Consultation. Definition, diagnosis and classification of diabetes mellitus and its complications. Part 1: diagnosis and classification of diabetes mellitus provisional report of a WHO consultation. Diabet Med. 1998;15:539–553. [PubMed]

[12]. Schafer AI. The hypercoagulable states. Ann Intern Med. 1985;102:814–828. [PubMed]

[13]. Zhou B. Coorperative Meta-Analysis Group Of China Obesity Task Force Predictive values of body mass index and waist circumference to risk factors of related diseases in Chinese adult population. Zhonghua Liu Xing Bing Xue Za Zhi. 2002;23:5–10. [PubMed]

[14]. Clinical and Laboratory Standards Institute. How to define and determine reference intervals in the clinical laboratory Approved Guideline-second Edition. 2000;20:C28–A2.

[15]. Carr ME. Diabetes mellitus: a hypercoagulable state. J Diabetes Complications. 2001;15:44–54.[PubMed]

[16]. Barazzoni R, Zanetti M, Davanzo G, Kiwanuka E, Carraro P, et al. Increased fibrinogen production in type 2 diabetic patients without detectable vascular complications: correlation with plasma glucagon concentrations. J Clin Endocrinol Metab. 2000;85:3121–3125. [PubMed]

[17]. Ten Boekel E, Bartels P. Abnormally short activated partial thromboplastin times are related to elevated plasma levels of TAT, F1+2, D-dimer and FVIII:C. Pathophysiol Haemost Thromb. 2002;32:137–142.[PubMed]

[18]. Lippi G, Salvagno GL, Ippolito L, Franchini M, Favaloro EJ. Shortened activated partial thromboplastin time: causes and management. Blood Coagul Fibrinolysis. 2010;21:459–463. [PubMed]

[19]. Mina A, Favaloro EJ, Mohammed S, Koutts J. A laboratory evaluation into the short activated partial thromboplastin time. Blood Coagul Fibrinolysis. 2010;21:152–157. [PubMed]

[20]. Reverter JL, Reverter JC, Tàssies D, Rius F, Monteagudo J, *et al*. Thrombomodulin and induced tissue factor expression on monocytes as markers of diabetic microangiopathy: a prospective study on hemostasis and lipoproteins in insulin-dependent diabetes mellitus. Am J Hematol. 1997;56:93–99. [PubMed]

[21]. Madi AM, Greci LS, Nawaz H, Katz DL. The activated partial thromboplastin time in early diagnosis of myocardial infarction. Blood Coagul Fibrinolysis. 2001;12:495–499. [PubMed]

[22]. Mostafa SA, Davies MJ, Srinivasan BT, Carey ME, Webb D, *et al*. Should glycated haemoglobin (HbA1c) be used to detect people with type 2 diabetes mellitus and impaired glucose regulation? Postgrad Med J. 2010;86:656–662. [PubMed]

[23]. Avignon A, Radauceanu A, Monnier L. Nonfasting plasma glucose is a better marker of diabetic control than fasting plasma glucose in type 2 diabetes. Diabetes Care. 1997;20:1822–1826. [PubMed]

[24]. Festa A, D'Agostino R, Tracy R, Haffner S. Elevated levels of acute-phase proteins and plasminogen activator inhibitor-1 predict the development of type 2 diabetes. Diabetes. 2002;51:1131–1137. [PubMed]

[25]. Kannel WB, D'Agostino RB, Wilson PW, Belanger AJ, Gagnon DR. Diabetes, fibrinogen and risk of cardiovascular disease: the Framingham experience. Am Heart J. 1990;120:672–676. [PubMed]

[26]. Van Wersch JW, Westerhuis LW, Venekamp WJ. Coagulation activation in diabetes mellitus.Haemostasis. 1990;20:263–9. [PubMed]

[27]. Missov R, Stolk R, van der Bom J, Hofman A, Bots ML, *et al*. Plasma fibrinogen in NIDDM: the Rotterdam Study. Diabetes Care. 1996;19:157–159. [PubMed]

[28]. Acang N, Jalil FD. Hypercoagulation in diabetes mellitus. Southeast Asian J Trop Med Public Health. 1993;24(Suppl 1):263–266. [PubMed]

[29]. Korte W, Clarke S, Lefkowitz JB. Short activated partial thromboplastin times are related to increased thrombin generation and an increased risk for thromboembolism. Am J Clin Pathol. 2000;113:123–127.[PubMed]

[30]. Ceriello A. Coagulation activation in diabetes mellitus: the role of hyperglycaemia and therapeutic prospects. Diabetologia. 1993;36:1119–1125. [PubMed]

[31]. Alao OO, Adebisi SI, Jombo GTA, Joseph DE, Damulak OD, *et al*. Cardiovascular Risk Factors among Diabetic Patients Attending a Nigerian Teaching Hospital. The Internet Journal of Endocrinology. 2010;6:122–132.