

Hemostasis in sickle cell disease

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Abstract

Sickle cell disease results in a significant morbidity and mortality related to intra-vascular thrombosis. This study is an attempt to improve our understanding the role of hypercoagulability in the pathogenesis of sickle cell disease. A case group of 20 asymptomatic sickle cell adult patients in a steady-state were compared with a control group of 20 normal adult people, both groups aged (18-50) year and from both sex, to evaluate the process of hemostasis. An investigation has been done for both groups including estimation of hemoglobin, platelets count, bleeding time and clotting time. There is a highly significant increase in the number of platelets with ($P < 0.01$) together with a highly significant decrease in bleeding time with ($P < 0.01$) for a case group in comparison with a control group. There is no significant difference of clotting time between both groups. That may suggest that the platelets aggregation formation activity is significantly increase in patients with sickle cell disease in a steady-state.

Keyword: Sickle cell disease, Platelets, Bleeding time

Introduction

Sickle cell disease (SCD) is an inherited lifelong disorder of genetic abnormality¹. Characterized by sickle (crescent)-shaped red blood cells and chronic anemia caused by excessive destruction of the abnormal red blood cells and multi-organ morbidity.² There is increasing evidence that SCD are usually associated with a hypercoagulable state and the risk of thrombotic complications appears to be higher in SCD patient.³

Hemostasis is a process of prevention the blood loss after vascular injury achieved by formation of clot,⁴ which is a mass formed from platelets and various blood proteins (clotting factors) within a blood vessels to stop bleeding.⁵

Platelets are tiny cell fragments that circulate in the blood stream and they are the first cells to react to blood vessels injury and seal off the wound, preventing more blood from escaping⁶.

The present study was designed to evaluate the platelets plug formation activity by measuring the bleeding time (BT) which is a standard test and basic assessment of how the blood platelets work to form a plug and stop bleeding, normal value is (2-7mints) for unisex and for all age groups.⁷ Also we measured

the clotting time (CT), which is the time required for a sample of blood to coagulate in vitro, the normal value is (8-15mints) for unisex and for all age group.⁸ By this test we can evaluate the ability of different clotting factors work to form a clot normally.

Materials and Method

Case group of a total 20 steady-state SCD patients from both sex, aged between (18-50) years old, recruited from Al-Sadder teaching hospital outpatient medical clinic, documented as homozygous for HbS using hemoglobin electrophoresis. Compared with 20 healthy control group from both sex and have the same age. Investigations has been done for both groups including hemoglobin concentration, platelet count, bleeding time and clotting time.

Bleeding time procedure

Pricking a sterilized finger about 4mm depth, at the same time press the stop watch, after 30 sec. we remove the blood ooze from the finger by a filter paper every 30 sec. until the miner bleeding disappear⁹.

Bleeding time is consider as the number of blood spots $\div 2$

Clotting time procedure

Pricking a sterilized finger about 4mm depth, filling a capillary tube with blood, at the same time press the stop watch and start to break down the capillary tube every 30 sec. looking for the formation of fibrin thread⁹.

Results and Discussion

The study was in a completely randomized method and the results performed with SPSS statistical software version 20 ANOVA analysis of variation.

Probability value of (P<0.05) considered to be statistically significant.

Table 1: Visualize the mean ± SD of hemoglobin concentration and platelets count

Parameters	Case	Control
	Mean ± SD	Mean ± SD
Hemoglobin Concentration	9.96 ± 1.216	11.54 ± 2.84
Platelets Count	402.37 ± 44.08	216.00± 5.03

***Statistically highly significant concenter as (P<0.01)**

The comparison between SCD adult patients with normal people shows highly significant increase in platelets count with highly significant decrease in bleeding time and hemoglobin concentration (P<0.01), as visualized in table(1) and table(2).

Table 2: Visualize the mean ± SD of Bleeding time & Clotting time

Parameters	Case	Control
	Mean ± SD	Mean ± SD
Bleeding Time	1.82 ± 0.96	3.40± 1.48
Clotting Time	2.95 ± 1.82	3.38 ± 1.50

***Statistically highly significant decrease of bleeding time (P<0.01)**

no significant differences for clotting time.

Sickle cell disease refers to a group of genetic disorders defined by the presence of sickle hemoglobin (HbS) that passed from parents to the children.¹⁰ This study is an attempt to improve our understanding the role of hypercoagulability in the pathogenesis of SCD.

The raised platelets count has previously been reported.¹¹ Platelets stimulation and chronic activation of coagulation is commonly observed in patient with SCD at steady-state compared to healthy control subjects with normal hemoglobin.¹¹ This is evidenced by increased plasma levels of *in vivo* markers of fibrin generation and thrombin, including fibrinopeptide A, D-dimers and plasmin-antiplasmin complex.¹² One of the most important factor that contribute to platelets

activation in SCD, is the abundance of free hemoglobin in the circulation, which is responsible for the alteration of platelets functions by limiting the bioavailability of nitric oxide (NO).¹³ It has been reported that NO play very important role to inhibit platelets aggregations and adhesion to subendothelium matrices/endothelium through the cyclic guanosine monophosphate (GMP) pathway.¹⁴ Furthermore, circulating HbS can potentiate the platelets activation and promote the platelet-thrombus formation on subendothelium matrix, also it can bound to glucoprotein (GPIIb) and activated the intracellular proteins of Lyn_ERK pathway in the platelets.¹⁵ Increased plasma level of markers of platelets in SCD patients are capable of stimulating endothelial cell activation as demonstrated by the induction of the increased expression of endothelial inflammatory and adhesive proteins.¹⁶ Furthermore, hyper activity of

platelets also found to be more vigorous in SCD painful crises that may suggest a possible therapeutic role of drugs to inhibit the platelets functions.¹⁷

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Conflict of Interest: None to declare.

Ethical Clearance: All experimental protocols were approved under the Physiology department / college of medicine/ University of Basrah, Iraq and all experiments were carried out in accordance with approved guidelines.

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