Review Articles

Sickle Cell Disease and Venous Thromboembolism

Zohreh Rahimi1,2 and Abbas Parsian3

1 Medical Biology Research Center, Kermanshah University of Medical Sciences, Kermanshah, Iran
2 Department of Biochemistry, Medical School, Kermanshah University of Medical Sciences, Kermanshah, Iran
3 Division of Neuroscience & Behavior, NIAAA, National Institutes of Health, Rockville, Maryland, USA

Correspondence to: Zohreh Rahimi, Ph.D. Associate Professor of Biochemistry, Medical Biology Research Center, Medical School, Daneshgah Avenue, Kermanshah, Iran P.O.Box: 67148-69914. Tel: 0098-831-4274618-21. Fax: 0098-831-4276471. E-mail: zrahimi@kums.ac.ir rahimizus@yahoo.com

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Abstract: Hemoglobin S in homozygous state or in combination with one of the structural variants of Hb D-Punjab, Hb O-Arab, Hb C or β-thalassemia mutation results in sickle cell disease (SCD) that is characterized by chronic hemolytic anemia and tissue injury secondary to vasoocclusion. A chronic hypercoagulable state in SCD has been established with the increased risk of thromboembolic complications in these patients. The goal of present review is to survey of the literature related to thromboembolic events and genetic risk factors involved in the manifestation of these events in SCD patients with focus on studies from Mediterranean countries. Also, this review covers the pathogenesis of hypercoagulability and alteration in the components of hemostasis system.

Introduction: The Hb S that results from substitution of valine for glutamic acid at position 6 of β-globin chain was the first hemoglobin variant to be discovered.1 This abnormal hemoglobin is the most clinically important structural variant of hemoglobin with highest frequency in Africa, Saudi Arabia, and India.1,2 Sickle cell disease (SCD) results from the homozygous state of the mutation, or a compound heterozygous state with one of structural variants of Hb D-Punjab, Hb O-Arab, Hb C or β-thalassemia mutation in the other β-gene.2 This group of disorders characterized by the polymerization of deoxygenated hemoglobin S into rigid rodlike polymers, causing the sickling of the erythrocyte.3

The β8 mutation has been found to be in linkage disequilibrium, with five distinct, typical (common) β-globin gene cluster haplotypes. Four of these haplotypes are known as African haplotypes (Bantu, Benin, Senegal, and Cameroon), the fifth is the Arab-Indian haplotype, which was originally described in the eastern oases of Saudi Arabia and among the tribal Adivasi population of India.4 The clinical severity and hematological manifestations of sickle cell anemia are varied and are influenced by other pleiotropic effects of the haplotype background linked to the β8 gene.5,6

SCD is known as a hypercoagulable state in which various hemostatic systems both in steady state and during vaso-occlusion are perturbed with increased
activation of the coagulation system and platelets, thrombin generation, and occurrence of thrombosis.\textsuperscript{7-10}

**Cerebral venous and sinus thrombosis (CVST):** The most important cause of stroke in SCD patients is large-vessel cerebral arterial complication that might be the result of abnormal adhesive and procoagulant properties of RBC, which produce endothelial damage, secondary intimal proliferation, and thrombosis.\textsuperscript{16-12}

The incidence of cerebro-vascular complications in SCD is 5 to 10\% (13). In 11\% of sickle cell anemia patients stroke is occurred by age 20, with infarction mainly in the internal carotid and middle cerebral arteries.\textsuperscript{14}

Association between SCD and cerebral thromboembolic events has been investigated in several studies. Alli et al\textsuperscript{15} reported the presence of skull bone infarction crisis and deep vein thrombosis in a boy with homozygous sickle cell anemia (SCA). Also, in individuals with sickle cell trait (SCT) cerebral infarction and stroke is not rare and there are several case reports related to the incidence of stroke in these individuals.\textsuperscript{16-19} There are few studies from Mediterranean countries reported the occurrence of CVST and stroke in SCD patients. Sidani et al\textsuperscript{20} have reported the existence of venous and sinus thrombosis and stroke in a Lebanese young homozygous sickle cell patient. However, in this patient the presence of proteins C, S, antithrombin III, factor V Leiden (FVL), and prothrombin mutations as well as anticardiolipin antibodies were negative.\textsuperscript{20} Also, a 25-year-old French SCD patient with cerebral vein thrombosis and bilateral thalamic infarcts has been reported.\textsuperscript{21} Further, the report of Ozu et al\textsuperscript{22} indicated the occurrence of sinus thrombosis and thalamic infarcts in a 2-year-old child from Turkey. In addition, Radhakrishnan et al\textsuperscript{16} reported the presence of stroke in two young adult sickle cell trait individuals from Libya.

**Pulmonary embolism and deep vein thrombosis:** A link between hypercoagulability in SCD patients and pulmonary hypertension (PHT), increased pulmonary artery pressure and pulmonary vascular resistance, has been established.\textsuperscript{2,23} Pulmonary hypertension occurs in approximately 30\% of adult patients with SCD and is a risk factor for early death.\textsuperscript{24} In studies using echocardiograms of patients with SCA who were examined at referral centers the prevalence of PHT was 20 to 30\%. In autopsy studies, approximately 75\% of patients with SCA present histological evidence of PHT.\textsuperscript{25}

Hemolysis has been identified as one of the potential driving forces of the hypercoagulable state and developing PHT in SCD.\textsuperscript{23,26} However, Beers et al\textsuperscript{27} did not detect a significant difference in haemolysis between patients with and without PHT and did not find a direct link of the hypercoagulable state to mild SCD related PHT.

A mechanism related to nitric oxide (NO) scavenging by free hemoglobin has been implicated in the pulmonary arterial disease of sickle cell anemia. Hemolytic anemia through hemolysis and release of hemoglobin into plasma which consumes NO and release arginase into plasma leads to resistance to NO-dependent vasodilatory effects. This is an important process in the development of PHT and shortened life expectancy in patients with SCD.\textsuperscript{25,28} Bunn et al\textsuperscript{29} reviewed the literature related to the influence of NO depletion by plasma hemoglobin in the microcirculation on the pathogenesis of PHT in SCD patients. They concluded that pulmonary hypertension per se was not a major cause of death in sickle cell patients and therapies that might enhance the availability of NO to the vasculature have thus far been ineffective and/or toxic.

In a large case-control study on 515 hospitalized black patients with thromboembolism and Hb S and C and 555 black controls revealed that persons with SCT experienced approximately a 2-fold increased risk of venous thromboembolism (VTE) compared with persons with the wild-type genotype, Hb AA. This increased risk was observed both for idiopathic and provoked VTE as well as for first and recurrent VTE. In addition, pulmonary embolism (PE) risk (without DVT) was significantly increased (approximately 4-fold) among those with SCT, whereas the risk of a DVT (without PE) was not significantly increased.\textsuperscript{30} The increased risk of VTE is attributed to subclinical sickling of red cells.\textsuperscript{30}

Results of autopsy of 12 SCD patients suggested that pulmonary thromboemboli may be a late complication of PHT and an in situ thrombotic arteriopathy underlies the development of PHT in most patients with sickle cell disease.\textsuperscript{29} Small vessel thrombosis is one of underlying cause of pulmonary hypertension (PHT) among patients with SCD (31) and an association between thromboembolism and PHT has been suggested.\textsuperscript{32} Staser et al\textsuperscript{33} reported a rare phenomenon of calcified pulmonary thromboembolism in an African American boy with SCD.

Higher prevalence of pulmonary embolism in African American SCD patients below 40 years of age (0.44\%) compared to non SCD African Americans (0.12\%) has been reported.\textsuperscript{34} Clinical assessment and/or autopsy findings at the time of death among 141 adults with sickle cell disease (SCD) over a 25-year period revealed that PHT is the leading cause of death (26.2\%) and thromboembolism was the fifth symptom leading to death in these patients. In another study with the goal of determining the most common pathologic
findings in autopsy cases with sickle cell lung disease indicated the high percentage of PHT (33.3%) in these autopsy samples. The risk of thromboembolic complications in SCD patients appears to be higher following splenectomy. Genetic of Thromboembolism: Venous thromboembolism is believed to be caused by genetic and acquired risk factors. The inherited hypercoagulable syndromes primarily affect veins, and only rarely cause arterial thrombosis. The acquired hypercoagulable states, such as the antiphospholipid antibody syndrome, are more commonly implicated in arterial stroke. The role of inherited thrombophilia in the pathogenesis of sickle cell thromboses has been reported in few studies. In these studies the frequency of thrombophilic mutations of FVL, prothrombin G20210A, and methylenetetrahydrofolate reductase (MTHFR) C677T and their association with incidence, and recurrence of thromboembolism among SCD patients have been examined. The factor V Leiden mutation is caused by a single point mutation at nucleotide 1691, leading to an Arg/Gln amino acid exchange at this position is the most frequent inherited risk factor for thromboembolism in Caucasians with a prevalence of 5% in the general population. Higher frequency of this mutation in β-thalassemia patients and its association with deep venous thrombosis and cerebral venous and sinus thrombosis has been reported. There are many studies reported the low frequency of thrombophilic mutations in SCD patients and the lack of association between these mutations and risk of thromboembolism. Helley et al. in a large sample of SCD patients from Africa indicated the absence of FVL mutation in these patients. However, the gene frequency of the FVL in black SCD subjects from West Indies and Maghrib were 2.5 and 1.1%, respectively. They concluded that in sub-Saharan African SCD patients FVL is not an additional risk factor for thrombosis. Also, In African American with SCD this mutation was uncommon and was not found to be responsible for stroke in these patients. Further, in the study of Wright et al. among SCD patients from Jamaica no FVL mutation was found in these patients. However, a significant reduction in the median activated protein C resistance ratio compared to controls was observed in their study. Moreira Neto et al. reported no prothrombin G20210A mutation in SCD patients from Brazil. The frequency of heterozygous FVL mutation was 1.8% in their studied patients. In another study from Brazil, Andrade et al. studied the prevalence of the FVL mutation, MTHFR C677T polymorphism, and prothrombin gene variant in SCD patients. It was reported no significant difference between SCD patients and the control population for the prevalence of the studied thrombophilic mutations. However, among nine studied SCD patients with vascular complications of stroke or deep vein thrombosis they found only one patient to be a carrier for FVL. Also, Kordes et al. reported no medical history of deep vein thrombosis in two SCA patients homozygous for FVL mutation from Iraq. Fawaz et al. have compared the prevalence of FVL mutation and prothrombin gene variant in SCD patients and controls from Eastern Saudi Arabia. Their study revealed that there is no association between the mutations and SCD. Furthermore, Zimmerman et al. reported no association between thrombophilic mutations of MTHFR C677T and platelet glycoprotein IIIa (GPIIIa) C1567T with SCD. In contrast, in spite of the moderate prevalence of FVL mutation (2.97-5.5%) among normal population of Iran a high prevalence of FVL mutation (14.3%) among Iranian SCA patients has been found with a significant association between this mutation and SCA with odds ratios (OR) of 6.5 (95% confidence intervals [CI] 1.19–35.33, p = 0.03). In addition, increased prevalence of the FVL mutation in SCT individuals and sickle/β-thalassemia patients was not statistically different from controls (OR=3.84, 95% CI 0.49–29.9, p = 0.19 and OR=3.77, 95% CI 0.31– 45.9, p = 0.29, respectively). Also, in a study from Brazil it was suggested that MTHFR C677T might be a risk factor for vascular complications in SCD. According to the literature, there have been three studies of inherited risk factors of venous thromboembolism in SCD patients from Southern Mediterranean countries which report high prevalence of thrombophilic mutations in SCD patients and their association with thromboembolism in these patients. Among Lebanese sickle/β0-thalassemia patients, a high prevalence of thrombophilic mutations of FVL (42%), homozygous and heterozygous MTHFR C677T (59%), and prothrombin G20210A (8%) has been reported. In this report sickle-β-thalassemia patients were 5.24 and 4.39 times more likely to have FVL mutation as compared to the normal controls and thalassemia intermedia patients, respectively (p <0.05). Also, the presence of extensive large vessel thrombosis in a sickle/β0-thalassemia patient from Lebanon doubles heterozygous for FVL and MTHFR C677T (homozygous for FVL and heterozygous for MTHFR has been reported. Further, in a sickle cell anemia patient from Israel, Koren et al. reported the recurrent of cerebrovascular accident and deep venous thrombosis. Activated protein C resistance due to FVL heterozygous and heterozygocity for the MTHFR C677T have been diagnosed and
suspected to be the risk factors that contribute to the development of the deep vein thrombosis in this SCA patient.

**Pathogenesis of Hypercoagulability:** The pathogenesis of hypercoagulability is considered to be multifactorial. Altered components of hemostasis system in SCD have been suggested. Low plasma levels of protein C, protein S, and antithrombin III, elevated plasma levels of thrombin-antithrombin (TAT) complexes, prothrombin fragment 1+2 (F1+2), D-dimer complexes, and circulating antiphospholipid antibodies, platelet activation during vaso-occlusive crisis, abnormal external exposure of phosphatidylserine (PS) and adherence of sickle erythrocytes to the vascular endothelium, reducing NO phosphatidylethanolamine, are exposed to external membrane of red blood cells (RBC) and permanently fixed in irreversibly sickled cells leading to promote coagulation of red blood cells and platelets. The enzyme of scramblase in a Ca\(^{++}\) dependent process which is modified by sulphydryl is involved in the translocation of PS from inner to outer layer of membrane. The increased oxidative stress in hemoglobinopathies increases sulphydryl modification and external exposure of PS. This process is a signal for RBC removal in apoptosis and a docking site for proteins involved in coagulation processes. Altered structure of RBC membrane in sickle cell disease due to external exposure of PS in the outer membrane increases adhesion of sickle erythrocytes to endothelial cells and microvascular occlusion. This alteration decreases red blood cell survival by increased splenic clearance of PS positive erythrocytes. Annexin V through calcium dependent mechanism binds to anionic phospholipids (primarily PS) exposed at external membrane layer. A high level of normal binding of annexin V to erythrocytes in sickle cell disease has been demonstrated. Painful vasoocclusive crises may partly result from these abnormal adhesive RBCs interacting with adhesion receptors on endothelia. Setty et al revealed a significant positive correlation between PS positive RBC and adhesion to vascular endothelium.

In SCD patients the presence of Arab-Indian and Senegal haplotype backgrounds of β gene compared to African haplotypes of Benin, Bantu and Cameroon are associated with higher levels of Hb F and amelioration of anemia. The benefit of Hb F effect in SCD patients has been shown that higher levels of F cells have a concomitant decrease in the numbers of microvesicles and PS-positive cells. The association between in vivo PS positive RBC and thrombin generation in SCD patients has been demonstrated by Setty et al. Westerman et al study indicated that phosphatidylserine positive RBC are a significant source of PS (+) vesicles in SCA and the levels of circulating RBC-derived vesicles are increased in SCA. They reported a linkage between vesicle levels and activation of thrombin. The correlation between subpopulations of PS (+) erythrocytes and the risk of stroke in patients with SCD has also been demonstrated. Overall, the role of RBC in the coagulation activation has been shown that includes a significant correlation between the numbers of RBCs with external exposure of phosphatidylserine with plasma markers of thrombin generation, such as prothrombin fragment 1+2 (F1+2), D-dimer and plasmin- antiplasmin complexes. However, such association was not detected between phosphatidylserine-positive platelets and markers of thrombin generation.
Elevation of Tissue Factor Expression: Tissue factor (TF) is a transmembrane glycoprotein that is involved in the activation of prothrombin and thrombin formation. Endothelial cells through expression of TF initiate coagulation. Significant elevation of TF in blood monocytes and circulating endothelial cells of SCD patients has been demonstrated that could be responsible for activation of the coagulation system in SCD patients.

The mechanisms of activation TF are 1) increased heme levels through hemolysis which induces TF expression on the surface of endothelial cells, ischemia-reperfusion injury (hypoxia/reoxygenation), 2) increased levels of soluble CD40 ligand through activation of platelets and exposure of CD40 ligand on their surface. In addition, the over expression of markers of endothelial activation such as endothelial adhesion proteins (intercellular adhesion molecule-1 [ICAM-1], E selectin [ELAM-1], P selectin and vascular cell adhesion molecule-1 [VCAM-1]) has been reported in SCD.

The utility of anticoagulant therapy as prophylaxis in hemolytic anemias such as SCD and novel approaches, including anti-hemolytic therapies, hemoglobin scavengers and NO donors have been suggested that could decrease the occurrence of thrombotic complications.

Conclusions: There are several evidences that prove the multifactorial pathogenesis of hypercoagulability in SCD that include disturbing red blood cell phospholipids membrane asymmetry, abnormal activation of endothelial cells and other blood cells. The hypercoagulable state in SCD patients could be more complicated in the presence of inherited and acquired risk factors of thrombosis. Future studies on the influence of thrombotic risk factors on the incidence of thromboembolic events in SCD patients could elucidate the actual risk of thromboembolism in these patients.

References:

1. Weatherall DJ, Glegg JB, Higgs DR, Wood WG. The hemoglobinopathies. In: Srivier CR, Benett AL, Sly WS, Valle D eds. Metabolic and Molecular Basis of Inherited Disease. New York: McGraw-Hill, Inc., 1995; Vol 3 pp. 3417-3484
2. Old JM. Hemoglobinopathies. In: Elles R, ed. Methods in Molecular Medicine: Molecular Diagnosis of Genetic Disease. Totowa, Humana Press Inc., 1996;169-183 doi:10.1385/JMB:1989090603-5:169 PMid:21374517
3. Bolanos-Meade J, Keung YK López-Arvizu C, Florendo R, Cobos E. Thrombotic thrombocytopenic purpura in a patient with sickle cell disease. Ann Hematol 1999; 78: 558-559 doi:10.1007/s002770050558
4. Rahimi Z, Merat A, Gerard N, Krishnamoorthy R, Nagel RL. Implications of the genetic epidemiology of globin haplotypes linked to the sickle gene in Southern Iran. Hum Biol. 2006; 78:719-731 doi:10.1353/hub.2007.0016 PMid:17564250
5. Rahimi Z, Karimi M, Haghshenass M, Merat A. Beta-globin gene cluster haplotypes in sickle cell patients from Southwest Iran. Ann Hematol 2003; 74:156-160 doi:10.1006/ajh.2002.8672 PMid:14587041
6. Niranjay, Y Chandak GR, Veeraraju P, et al. Some atypical and rare sickle cell gene haplotypes in populations of Andhra Pradesh, India. Hum Biol 1999; 71:333-340 PMid:10380370
7. Ataga KI, Cappellini MD, Rachmilewitz EA. Beta-thalassemia and sickle cell anaemia as paradigms of hypercoagulability. Br J Haematol 2007; 139: 3-13 doi:10.1111/j.1365-2141.2007.06740.x PMid:17854302
8. Ataga KI, Orlinger EP. Hypercoagulability in sickle cell disease: a curious paradox. Am J Med 2003;115: 721–728 doi:10.1016/j.amjmed.2003.07.011 PMid:14693325
9. Rahimi Z, Vaisi-Raygani A, Nagel RL, Muniz A. Thrombophilic mutations among Southern Iranian patients with sickle cell disease: high prevalence of factor V Leiden. J Thromb Thrombolysis 2008, 25: 288-292 doi:10.1007/s11239-007-0069-x PMid:17619828
10. Francis RB. Large-vessel occlusion in sickle cell disease: pathogenesis, clinical consequences, and therapeutic implications. Med Hypotheses 1991; 35: 88-95 doi:10.1016/0306-9877(91)90029-X
11. Francis RB Jr. Platelets, coagulation, and fibrinolysis in sickle cell disease: their possible role in vascular occlusion. Blood Coagul Fibrinolysis 1991; 2: 341-353 doi:10.1097/000001721-199104000-00018
12. Francis RB, Johnson CS. Vascular occlusion in sickle cell disease: current concepts and unanswered questions. Blood 1991; 77: 1405-1414 PMid:2009364
13. Helley D, Besmond C, Ouscroq R, da Silva F, Guillin MC, Bezaud E, Elion J. Polymorphism in exon 10 of the human coagulation factor V genein a population at risk for sickle cell disease. Hum Genet 1997; 100: 245-248 doi:10.1007/s004390050499 PMid:9254858
14. Ouhene-Frempong K, Weiner SJ, Sleeper LA. Cerebrovascular accidents in sickle cell disease: rates and risk factors. Blood 1998; 91: 288-94 PMid:9414296
15. Alli NA, Wainwright RD, Mackinnon D, Poyiadjiis S, Naida G. Skull bone infarctive crisis and deep vein thrombosis in homozygous sickle cell disease: case report and review of the literature. Hematology 2007; 12, 169-174 doi:10.1080/10245330601111912 PMid:17454200
16. Radhakrishnan K, Thacker AK, Maloo JC, Mangouss MA. Sickle cell trait and stroke in the young adult. Postgrad Med J 1990; 66, 1078 – 1080 doi:10.1136/pgmj.66.782.1078 PMid:2084660 PMCID:2429781
17. Handler CE, Perkin, GD. Sickle cell trait and multiple cerebral infarctions. J R Soc Med 1982, 75: 550-553 PMid:7086897 PMCID:1437918
18. Greenberg J, Massey EW. Cerebral infarction in sickle cell trait. Ann Neurol 1985, 18: 354-355 doi:10.1002/ana.410180315 PMid:4051462
19. Reyes MG. Cerebrovascular infarctions in sickle cell trait. J Neurol Neurosurg Psychiatry 1989, 52, 516-518 doi:10.1136/insp.52.4.516 PMid:50411
20. Sidani CA, Ballourah W, El Dassouki M, Muwakkit S, Dabbous I, Dahoui H, Al-Kutoubi A, Aboud MR. Venous sinus thrombosis leading to stroke in a patient with sickle cell disease on hydroxyurea and high hemoglobin levels: Treatment with thrombolysis. Am J Hematol 2008; 83: 818-820 doi:10.1002/ajh.21261 PMid:18756541
21. Di Roio C, Jourdan C, Terrier A, Artru F. Sickle cell anemia and internal cerebral vein thrombosis. Ann fr Anesth Reanim 1997; 16: 967-969 PMid:9750646

Med J Hematol Infect Dis 2011; 3: Open Journal System
Rahimi Z, Mozafari H, Amirigbigyan AH, Douibali RM, Vaisraygani A, Afsahi D, Razazzian N, Rezaei M. Cerebral venous and sinus thrombosis and thrombophilic mutations in Western Iran: association with factor V Leiden. Clin Appl Thromb Hemost. 2013; 19: 430-434. doi:10.1177/1076029209335519 PMid:19703820

Kahn MJ, Scher C, Rozans M, Michaels RK, Leissinger C, Krause J. Factor V Leiden is not responsible for stroke in patients with sickling disorders and is uncommon in African Americans with sickle cell disease. Am J Hematol 1997; 54: 12-15. doi:10.1002/(SICI)1096-8652(19970115)54:1<12::AID-AJH2-3.0.CO;2-7

Kahn JE, Veysseer-Belot C, Renier JL, de Mazancourt P, Peltier JY, de Raucourt E. Recurrent thromboembolism in a patient with β-thalassemia major associated with double heterozygosity for factor V R506Q and prothrombin G20210A mutations. Blood. 2003; 102: 1461-1463. doi:10.1182/blood-2002-07-200012

Wright JG, Cooper P, Malia RG, Kulozik AE, Vetter B, Thomas P, Preston FE, Serjent GR. Activated protein C resistance in homozygous sickle cell anemia. Br J Haematol 1997; 96: 854-856. doi:10.1046/j.1365-2141.1997.doi:10.1111/j.1365-2141.1997.doi:10.20843.spmid:9074431

Moreira Neto F, Lourenço DM, Noguti MAE, Morelli VM, Gil ICP, Beltrão ACS, Figueiredo MS. The clinical impact of MTHFR polymorphism on the vascular complications of sickle cell disease. Brazilian J Med Biol Res 2006; 39: 1291-1295. doi:10.1590/S1384-975320060001200015

Kordes U, Janka-Schaub G, Kulozik A, Schneppenheim R. Homozygous factor V Leiden mutation in sickle cell anemia. Br J Haematol 2002; 116: 236. doi:10.1046/j.1365-2141.2002.02342.x. PMid:11848090

Fawaz NA, Bashawery L, Al-Sheikh I, Qateri A, Al-Othman SS, Almasi WY. Factor V-Leiden, prothrombin G20210A, and MTHFR C677T mutations among patients with sickle cell disease in Brazil. Am J Hematol 1998; 59: 46-50. doi:10.1002/(SICI)1096-8652(199809)59:1<46::AID-AJH30.0.CO;2-Z

Zimmerman SA, Ware RE. Inherited DNA mutations contributing to thrombophilic complications in patients with sickle cell disease. Am J Hematol. 2000; 60: 267-272. doi:10.1002/(SICI)1096-8652(199812)59:4<267::AID-AJH130.0.CO;2-W

Rahimi Z, Vaisraygani A, Mozafari H, Kharrazi H, Rezaei M, Nabioglu PA. Prevalence of factor V Leiden in eastern Mediterranean sickle-beta-thalassemia patients. J Thromb Thrombolysis 2000; 9: 353-352. PMid:10739401

Ismaeel H, Shamsheddeen AW, Mahfouz R, Zeineh N, Jndri O, Taher A. Screening for inherited thrombophilia might be warranted among Eastern Mediterranean sickle-β-thalassemia patients. J Thromb Thrombolysis 2006; 22:121–123 PMid:1708978

Otrock ZK, Mahfouz RA, Taher AT. Should we screen Eastern Mediterranean sickle beta-thalassemia patients for inherited thrombophilia? J Thromb Haemost 2005; 3: 599-600 doi:10.1111/j.1538-7836.2005.01148.x. PMid:1574826

Koren A, Zalman I, Levin C, Abu Hana M, Marder R, Shalev S. Venous thromboembolism, factor V Leiden, and methylenetetrahydrofolate reductase in a sickle cell anemia patient. Pediatr Hematol Oncol 1999; 16: 469-472. doi:10.1080/0888001992770747

El-Hazmi MA, Warys AS, Bahukun H. Blood proteins C and S in sickle cell disease. Acta Haematol 1993; 90: 114-119. doi:10.1111/j.1120-5371.1993.tb03519.x. PMid:8291368
Mediterr J Hematol Infect Dis 2011; 3: Open Journal System

55. Liesner R, Mackie I, Cookson J, McDonald S, Chitlolie A, Donohoe S, Evans J, Hann I, Machin S. Prothrombic changes in children with sickle cell disease: relationships to cerebrovascular disease and transfusion. Br J Haematol 1998; 103: 1037-1044 doi:10.1046/j.1365-2141.1998.01121.x PMid:9886316.

56. Onwemelukwe GC, Jibril HB. Anti-thrombin III deficiency in Nigerian children with sickle cell disease: possible role in the cerebral syndrome. Trop Geogr Med 1992; 44: 37-41 PMid:1496720.

57. Bayazit AK, Kilinc Y. Natural coagulation inhibitors (protein C, protein S, antithrombin) in patients with sickle cell anemia in a steady state. Pediatr Int 2001; 43: 592-596 doi:10.1046/j.1442-200X.2001.01476.x PMid:1173735.

58. Baker JE, Wandersse NJ. Thrombosis in heritable hemolytic disorders. Curr Opin Hematol 1999; 6: 71-75 doi:10.1097/00062752-199903000-00003 PMid:1008635.

59. de Jong K, Kuypers FA. Sulphydryl modifications alter scramblase activity in murine sickle cell disease. Br J Haematol 2006; 133: 427-432 doi:10.1111/j.1365-2141.2006.06045.x PMid:16643451.

60. Hebbel, RP. Beyond hemoglobin polymerization: the red blood cell membrane and sickle disease pathophysiology. Blood, 1991; 77, 214–237 PMid:1985689.

61. Fadok VA, Voelker DR, Campbell PA, Cohen JJ, Bratton DL, Henson PM. Exposure of phosphatidylserine on the surface of apoptotic lymphocytes triggers specific recognition and removal by macrophages. J Immunol 1992; 148: 2207–2216. PMid:1545126.

62. Fadok VA, Bratton DL, Rose DM, Pearson A, Ezekewitz RAB, Henson PM. A receptor for phosphatidylserine-specific clearance of apoptotic cells. Nature 2000; 405: 85–90. doi:10.1038/35011084.

63. Martin SJ, Reutelingsperger CPM, McGahon AJ, Rader JA., Van Schie R, LaFace DM, Green DR. Early redistribution of plasma membrane phosphatidylserine is a general feature of apoptosis regardless of the initiating stimulus: Inhibition by over expression of Bcl-2 and Abi. J Exp Med 1995; 182: 1545–1556 doi:10.1084/jem.182.5.1545 PMid:7595224.

64. Zwaal RFA, Schroit AJ. Pathophysiologic implications of membrane phospholipid asymmetry in blood cells. Blood. 1997; 89; 1121–1132 PMid:9028933.

65. Wood BL, Gibson DF, Tait JF. Increased erythrocyte phosphatidylserine exposure in sickle cell disease: flow-cytometric measurement and clinical associations. Blood 1996; 88: 1873-1880 PMid:8781447.

66. Kaul DK, Fabry ME, Nagel RL. Microvascular sites and characteristics of sickle cell adhesion to vascular endothelium in shear flow conditions: pathophysiological implications. Proc Natl Acad Sci US A. 1989; 86: 3356–3360 doi:10.1073/pnas.86.9.3356.

67. Setty BNY, Kulkarni S, Stuart MJ. Role of erythrocyte phosphatidylserine in sickle red cell-endothelial adhesion. Blood 2002; 99: 1564-1571 doi:10.1182/blood.V99.5.1564 PMid:11861269.

68. Setty BN, Kulkarni S, Rao AK, Stuart MJ. Fetal hemoglobin in sickle cell disease: relationship to erythrocyte phosphatidylserine exposure and coagulation activation. Blood 2000; 96:1119-24 PMid:10910931.

69. Setty BN, Rao AK, Stuart MJ. Thrombophilia in sickle cell disease: the red cell connection. Blood 2001; 98: 3228–33. doi:10.1182/blood.V98.12.3228 PMid:1179353.

70. Westerman M, Pizziy A, Hirschman J, Cerino M, Weil-Weiner Y, Ramotar P, Ezee A, Lawrie A, Purdy G, Mackie I, Porter J. Microvesicles in haemoglobinopathies offer insights into mechanisms of hypercoagulability, haemolysis and the effects of therapy. Br J Haematol 2008; 142: 126-135 doi:10.1111/j.1365-2141.2008.07155.x PMid:1842994.

71. Styles L, de Jong K, Vichinsky E, Labin B, Adams R, Kuypers F. Increased RBC phosphatidylserine exposure in sickle cell disease patients at risk for stroke by transcranial doppler screening. Blood 1997; 90: 604a.

72. Key NS, Stungla A, Dandelet L, Nelson SC, Moertel C, Styles LA, Kuypers FA, Bach RR. Whole blood tissue factor procoagulant activity is elevated in patients with sickle cell disease. Blood1998; 91: 4216-4223 PMid:9596669.

73. Solovey A, Gui L, Key NS, Hebbel RP. Tissue factor expression by endothelial cells in sickle cell anemia. J Clin Invest 1998; 101: 1899-1904 doi:10.1172/JCI1932 PMid:9576754.

74. Solovey A, Kollander R, Shet A, Milbauer LC, Panoskaltsis-Mortari A, Blazar BR, Kelm RJ Jr, Hebbel RP. Endothelial cell expression of tissue factor in sickle mice is augmented by hypoxia/reoxygenation and inhibited by lovastatin. Blood 2004; 104:840-846 doi:10.1182/blood-2003-10-3719 PMid:15073034.

75. Lee SP, Ataga KI, Orringer EP, Phillips DR, Parise LV. Biologically active CD40 ligand is elevated in sickle cell anemia: potential role for platelet-mediated inflammation. Arterioscler Thromb Vasc Biol 2006;26:1626-1631 doi:10.1161/01.ATV.0000220374.00602.e2 PMid:16601237.

76. Eldar A, Rachmilewitz EA. The hypercoagulable state in thalassemia. Blood 2002; 99: 36-43 doi:10.1182/blood.V99.1.36 PMid:11756150.