Review Article *Iran J Ped Hematol Oncol. 2025, Vol 15, No 3, 564-581*

Beyond the Blockage: A Deep Dive into the Pathogenesis of Vaso-Occlusive Crisis in Sickle Cell Disease

Maedeh Mohammadi MD[1], Hakimeh Hadi MSc[2], Hassan Boustani PhD[3], Ehsan Kamali Yazdi MD[4], Vahid Goodarzi MD[5*]

1. Department of Internal Medicine, School of Medicine, Guilan University of Medical Sciences, Rasht, Iran.
2. Thalassemia & Hemoglobinopathy Research Center, Health Research Institute, Ahvaz Jundishapur University of Medical Sciences, Ahvaz, Iran
3. Department of Hematology, Paramedical School of Science, Ilam University of Medical Sciences, Ilam, Iran
4. Alborz University of Medical Sciences, Alborz, Iran
5. Department of Anesthesiology, Rasoul-Akram Medical Center, Iran University of Medical Sciences (IUMS), Tehran, Iran
*Corresponding author: Dr. Vahid Goodarzi, Department of Anesthesiology, Rasoul-Akram Medical Center, Iran University of Medical Sciences (IUMS), Tehran, Iran. Email: vahid396396@gmail.com, ORCID ID: 0009-0003-6749-9408.

Received: 13 July 2024 **Accepted:** 25 February 2025

Abstract

Vaso-occlusive crisis (VOC) is the hallmark and most debilitating complication of sickle cell disease (SCD), yet its pathophysiology is multifactorial and not completely understood. This review examines the molecular and cellular events that drive VOC, highlighting the synergistic interaction between hypoxia, inflammation, and coagulation. Recurrent deoxygenation triggers hemoglobin S polymerization, causing red blood cell (RBC) sickling, loss of deformability, and microvascular obstruction. These events lead to endothelial activation, leukocyte adhesion, and platelet-leukocyte aggregation, fostering a hyperinflammatory, prothrombotic milieu. The increase in oxidative stress and release of cytokines result in neutrophil extracellular trap (NET) formation, exacerbating vascular injury and sustaining thromboinflammation. Together, these processes form a self-perpetuating loop, where hypoxia-induced inflammation and immunothrombosis reinforce VOC onset and severity. By elucidating these interlinked pathways, the review highlights novel therapeutic targets, particularly those modulating endothelial dysfunction, platelet-neutrophil crosstalk, and NET-driven coagulopathy. These mechanistic insights open new avenues for targeted interventions aimed at disrupting the VOC cycle and improving clinical outcomes in SCD. VOC in SCD represents a complex, self-amplifying pathological cascade driven by the interconnected processes of hypoxia, inflammation, and coagulation. Hypoxia, initiated by microvascular occlusion and compounded by impaired hemoglobin oxygen delivery, triggers a systemic inflammatory response, mobilizing innate and adaptive immune cells that further damage the endothelium and perpetuate vascular obstruction. Concurrently, hypoxia-induced activation of ECs and platelets facilitates a hypercoagulable state, characterized by TF expression, NETosis, and impaired anticoagulant mechanisms. These thromboinflammatory events not only exacerbate local ischemia but also extend systemically, contributing to multi-organ dysfunction and long-term morbidity in SCD patients. This review aims to provide an in-depth analysis of the molecular and cellular mechanisms that underlie VOC in SCD, with particular emphasis on the roles of hypoxia, inflammation, and coagulation.

Keywords: Endothelial Cell, Hemoglobin S, Neutrophil extracellular Trap, Pathogenesis, Thrombosis

Introduction

Vaso-occlusive crisis (VOC) represents one of the most debilitating and defining clinical manifestations of sickle cell disease (SCD), characterized by episodes of acute pain, tissue ischemia, and organ damage (1, 2). Despite significant advancements in our understanding of SCD, the precise mechanisms underlying VOC remain multifactorial and complex (3). SCD is marked by the presence of hemoglobin S (Hb S), which polymerizes under conditions of low oxygen tension, leading to red blood cell (RBC) sickling, microvascular occlusion, and impaired blood flow (4). These changes initiate a cascade of pathological events involving endothelial dysfunction, inflammatory cell activation, and alterations in hemostatic balance.

[DOI: 10.18502/ijpho.v15i3.18922]

Among these processes, hypoxia serves as a primary driver of RBC sickling and endothelial injury (5, 6). The dynamic interaction between hypoxia and subsequent cellular responses, such as oxidative stress and inflammation, forms a vicious cycle that exacerbates VOC episodes (7). As sickled RBCs become trapped within the microcirculation, their interaction with endothelial cells (ECs), platelets, and leukocytes promotes a pro-inflammatory and prothrombotic environment, which further sustains the occlusion (8, 9). Additionally, the activation of neutrophils and the release of neutrophil extracellular traps (NETs) have emerged as significant contributors to the thromboinflammatory milieu, highlighting a complex network of interactions that worsen vascular damage and perpetuate the crisis (10).

This review aims to provide an in-depth analysis of the molecular and cellular mechanisms that underlie VOC in SCD, with particular emphasis on the roles of hypoxia, inflammation, and coagulation. By exploring these interconnected pathways, this article seeks to identify potential therapeutic targets to break the cycle of VOC and improve clinical outcomes for individuals with SCD.

Pathogenesis and Clinical Consequences of VOC

VOC is the most prevalent and clinically significant complication of SCD, responsible for an estimated 197,000 emergency department visits each year (11). These painful episodes manifest in various anatomical locations and forms, ranging from acute conditions such as dactylitis (hand-foot syndrome) and hepatic sequestration to chronic syndromes including osteomyelitis and neuropathic pain (11). VOC is triggered when hypoxic conditions, driven by hemoglobin abnormalities, increase the polymerization of Hb S, leading to RBC sickling, membrane injury, and premature hemolysis. The resulting release of free hemoglobin, labile iron, and reactive oxygen species (ROS) promotes oxidative stress and a pro-inflammatory vascular environment (12-16).

These changes activate ECs, leukocytes, and platelets, enhancing cellular adhesion and contributing to microvascular occlusion. Impaired tissue perfusion exacerbates ischemia-reperfusion injury, ultimately causing localized tissue damage and the hallmark pain associated with VOC (12-15). Repeated episodes contribute to progressive end-organ damage, especially in the kidneys, liver, and skeletal system, further deteriorating patients' quality of life (12).

Previously, VOC treatment focused on symptomatic relief using hydration, opioids, and nonsteroidal anti-inflammatory drugs (NSAIDs) (13).

Today, more advanced interventions such as hydroxyurea (to increase fetal hemoglobin levels), chronic transfusion therapy for high-risk patients, and hematopoietic stem cell transplantation are more commonly used (17). Promising future strategies aim to target VOC pathogenesis directly by reducing cell adhesion and inflammation, through the use of oxygen affinity modifiers and anti-inflammatory agents (17).

Molecular Mechanisms Underlying Hb S Polymerization

Hb S polymerization is the primary molecular event driving the pathogenesis of SCD (18). A point mutation in the β-globin gene replaces glutamic acid with valine, creating a hydrophobic site on deoxygenated Hb S. This change promotes hydrophobic interactions between valine and adjacent residues, alanine, phenylalanine, and leucine, on neighboring Hb S molecules, facilitating tetramer formation (18). These tetramers periodically aggregate into double-stranded polymers. Seven of these strands assemble laterally to form helical, 14-stranded fibers approximately 20 nm in diameter (19-22). Stabilization of these fibers occurs through axial and lateral contacts among tetramers. Polymerization

[DOI: 10.18502/ijpho.v15i3.18922]

begins with a delay phase, during which 15–30 tetramers nucleate to form a stable core, a process known as homogeneous nucleation. Subsequent heterogeneous nucleation involves the addition of new tetramers onto existing fibers, leading to rapid, exponential polymer growth. Once approximately 250 tetramers accumulate, dense, insoluble polymer networks form, deforming RBCs and impairing their deformability and flow dynamics (19-22).

The kinetics of polymerization are influenced by the degree of deoxygenation and the intracellular levels of Hb S and fetal hemoglobin (Hb F). Oxygenation inhibits the exposure of hydrophobic sites, preventing tetramer formation and destabilizing early polymers. Hb F, which lacks the hydrophobic domain necessary for valine binding, disrupts polymer formation, explaining the therapeutic benefit of Hb F-inducing agents such as hydroxyurea (23). Therapeutic strategies also target polymerization by increasing oxygen affinity, lowering Hb S concentration, or interfering with intermolecular contacts. Ultimately, the clinical severity of SCD is tightly linked to the kinetics of Hb S polymerization, which is amplified under hypoxic conditions and impeded blood flow, allowing sufficient delay time for fiber nucleation and growth (24) (Table I).

ECs and Their Role in VOC Pathogenesis

ECs, which form the innermost lining of blood vessels, act as a dynamic interface between circulating blood and vascular walls. These cells closely regulate vascular tone, hemostasis, immune surveillance, and inflammatory responses. In the context of SCD, endothelial activation or dysfunction serves as a central contributor to pathological events such as VOC and atherosclerosis (25).

The endothelial surface provides a conducive environment for the adhesion and accumulation of sickled RBCs. Activated ECs express a range of adhesion molecules, including selectins and integrins, which promote the attachment of both leukocytes and deformed RBCs. Moreover, ECs influence clot formation and vascular tone by releasing prothrombotic and vasoactive mediators, further promoting VOC episodes (26). SCD is characterized by a heterogeneous population of RBCs, some of which display membrane damage and reduced lifespan (27). These damaged cells exhibit a greater tendency to adhere to ECs and other blood components. The adhesion process initiates with P-selectin-mediated interactions, followed by stabilization through molecules such as VCAM-1, CD36, integrins, and thrombospondin-1 (TSP) (28). Injury to the endothelial monolayer exposes subendothelial matrix proteins, intensifying cell adhesion and obstructing blood flow (29). Hypoxia, hemolysis, and recurring VOC episodes exacerbate endothelial dysfunction, upregulating the expression of adhesion molecules and elevating oxidative stress. This, in turn, recruits monocytes and neutrophils via interactions with EC-expressed selectins and intercellular adhesion molecules (ICAMs), leading to leukocyte extravasation and enhanced inflammation. ECs also regulate platelet function in this cascade. Specifically, the binding of platelet CD47 to endothelial TSP activates α2β3 integrins, facilitating interactions with ICAM-4 and ICAM-1 through fibrinogen bridges, thereby worsening microvascular occlusion (30, 31). Under normal conditions, ECs maintain anticoagulant and anti-inflammatory states. In SCD, however, endothelial dysfunction skews this balance toward a prothrombotic phenotype. Proinflammatory signaling induces the secretion of granules containing von Willebrand factor (VWF) and factor VIII, promoting platelet aggregation and vascular blockage. Additionally, ECs express elevated levels of tissue factor (TF), driving thrombin generation. Once

[DOI: 10.18502/ijpho.v15i3.18922]

activated, thrombin interacts with protease-activated receptors (PARs) on ECs, perpetuating vascular injury (32). One of the key homeostatic roles of ECs is the synthesis of nitric oxide (NO), which inhibits vasoconstriction, platelet adhesion, leukocyte recruitment, and smooth muscle proliferation via the cyclic guanosine monophosphate (cGMP) signaling pathway. In SCD, endothelial NO synthesis is impaired due to reduced levels of essential cofactors including arginine and tetrahydrobiopterin (BH4), and increased levels of inhibitors such as asymmetric dimethylarginine (ADMA). Under these conditions, endothelial nitric oxide synthase (eNOS) becomes uncoupled, producing superoxide instead of NO, thereby contributing to oxidative stress and vascular constriction. Furthermore, hemolysis and inflammatory stimuli (e.g., TNF) increase endothelial arginase activity, depleting arginine levels and further impairing NO production. The interaction of superoxide with NO generates peroxynitrite, which oxidizes BH4 and compounds endothelial dysfunction (Figure 1) (33). Given the pivotal role of ECs in VOC development, recent therapeutic strategies have targeted endothelial adhesion pathways to reduce disease severity. In a Phase 2 clinical trial, Ataga et al. demonstrated that treatment with crizanlizumab, a monoclonal antibody against P-selectin, significantly lowered the frequency of VOC episodes by inhibiting RBC-EC adhesion (34). In addition, a Phase 3 trial involving rivipansel, an E-selectin inhibitor, showed that this agent could reduce VOC duration and the need for opioid analgesia in SCD patients by blocking leukocyte-endothelium interactions (Table II) (35).

Hypoxia and Its Implications in SCD-Related VOC

Patients with SCD, particularly those with homozygous SS genotype, often exhibit reduced oxygen saturation compared to healthy individuals. Oxygen partial pressure (PO_2) shows a direct correlation with Hb levels and Hb F expression and an inverse association with leukocyte counts (36). Dense RBCs, which are enriched in Hb S, are more prevalent under hypoxic conditions and contribute to oxygen desaturation, further promoting sickling and vascular occlusion (Figure 2) (37). A key contributor to desaturation is a rightward shift of the oxyhemoglobin dissociation curve (ODC), indicating reduced oxygen affinity of Hb and an elevated P_{50} value, the oxygen tension at which hemoglobin is half-saturated. This shift is amplified in SCD due to both the intrinsic properties of Hb S and elevated levels of 2,3-diphosphoglycerate (2,3-DPG), a metabolic byproduct upregulated in response to chronic anemia and hypoxia. These adaptations facilitate oxygen unloading to tissues but inadvertently exacerbate hypoxia by destabilizing oxygen transport (38). Hemolysis in SCD also elevates circulating levels of dyshemoglobins, such as carboxyhemoglobin (COHb) and methemoglobin (MetHb), which are incapable of effectively binding or releasing oxygen. Their accumulation further compromises arterial oxygen content and contributes to systemic hypoxemia (39, 40). Anemia and morphological changes in sickled RBCs, which impair their deformability and reduce their circulation time, also diminish oxygen-carrying capacity and exacerbate tissue hypoxia (41, 42).

Endothelial dysfunction plays a central role in this process. Hemolysis releases free hemoglobin and ROS that rapidly scavenge NO, a vasodilator critical for maintaining vascular tone. The resultant NO depletion leads to vasoconstriction, impaired oxygen delivery, and increased cellular adhesion within the vasculature. Moreover, reduced NO availability enhances the adhesiveness of sickled cells, fostering an environment conducive to vaso-occlusive events (43). Therapeutic interest in restoring NO bioavailability has led to studies exploring the efficacy of

[DOI: 10.18502/ijpho.v15i3.18922]

inhaled NO in SCD. Although preliminary findings and animal models suggested beneficial effects, including case reports and early trials, a subsequent Phase 2 clinical study failed to demonstrate significant improvements in VOC duration or clinical outcomes, highlighting the complexity of NO-based interventions (44-47). VOC episodes may also cause upper airway obstruction and hypoventilation, further impairing oxygen exchange and exacerbating desaturation. This initiates a vicious cycle, hypoxia contributes to further Hb S polymerization and hemolysis, which in turn increases ATP release. Extracellular ATP is degraded into adenosine, which, through the engagement of A2B adenosine receptors (ADORA2B), enhances 2,3-DPG synthesis and perpetuates sickling (41). Additionally, hypoxia stabilizes hypoxia-inducible factors (HIFs), particularly HIF-1α, which promote extracellular adenosine accumulation, suppress eNOS activity, and amplify vascular injury. Nonetheless, HIF-1α also induces protective pathways, such as the upregulation of heme oxygenase-1 (HO-1), which facilitates heme degradation and may mitigate hemolysis-induced damage (48, 49).

Experimental models further demonstrate that hypoxic stress increases plasma levels of scavenging proteins such as hemopexin (Hx) and haptoglobin (Hp), which bind free hemoglobin and heme, thereby attenuating hemolysis-associated toxicity (50).

Ultimately, hypoxia serves as both a trigger and a consequence of vaso-occlusion. The sequence begins with reduced oxygen delivery, triggering sickling and hemolysis. This, in turn, promotes vascular adhesion and occlusion, leading to VOC and tissue ischemia. The resulting hypoxia perpetuates this cycle, causing severe pain and systemic complications (51). Despite the potential benefits of oxygen therapy in reversing early sickling events, clinical outcomes remain inconsistent. Notably, oxygen supplementation does not appear to significantly shorten VOC duration or alleviate pain severity, underscoring the need for further investigation into the complex role of hypoxia in SCD pathophysiology (52).

Inflammation in SCD

In SCD, persistent hemolysis, particularly the intravascular rupture of sickled RBCs, drives a potent inflammatory cascade. Approximately one-third of this hemolysis occurs within blood vessels, releasing free hemoglobin and heme into the bloodstream. When the body's binding proteins, such as haptoglobin and hemopexin, become overloaded, free hemoglobin binds to NO, effectively sequestering it and impairing endothelial function and vasodilation (53).

Simultaneously, the imbalance between ROS production and antioxidant defenses exacerbates endothelial damage (54). This oxidative stress originates from several sources, including Hb S autoxidation (55), elevated xanthine oxidase (XO) (56), NADPH oxidase (57), cytochrome P450, and cyclooxygenase activity (58). These oxidative damages result in EC dysfunction, triggering the expression of adhesion molecules and recruiting leukocytes and platelets to the site of injury.

Macrophages and dendritic cells within the tissues release interleukin (IL)-23, driving the production of IL-17A by T cells. This cascade stimulates the release of granulocyte colony-stimulating factor (G-CSF), promoting neutrophil activation and adhesion to the injured endothelium through chemokines and adhesion molecules. As hemolysis and tissue injury continue, damaged cells release danger-associated molecular patterns (DAMPs), including heme, ATP, mitochondrial DNA, and heat shock protein 70 (HSP70), which activate pattern recognition receptors (PRRs) such as toll-like receptors (TLRs) on ECs, macrophages,

[DOI: 10.18502/ijpho.v15i3.18922]

and platelets, triggering inflammasome formation and the release of pro-inflammatory cytokines (59-61).

The inflammasome, a cytoplasmic protein complex composed of adapter protein apoptosis-associated speck-like protein containing a CARD (ASC) and procaspase-1, plays a critical role in promoting inflammation. Once activated, caspase-1 processes IL-1β and IL-18 into their mature forms, amplifying acute inflammatory responses. This activation also triggers pyroptosis, a highly inflammatory form of cell death (62). In parallel, DAMPs stimulate neutrophils to release NETs, composed of chromatin and granule proteins, which entrap RBCs and platelets, further contributing to occlusion and vessel rigidity (63).

Platelets, in turn, act as amplifiers of the inflammatory response. They induce IL-8 production in ECs via nuclear factor kappa B (NF-κB) signaling and secrete cytokines, such as IL-1β, TNFSF14, and IL-6, which recruit neutrophils and exacerbate NET formation (64). Furthermore, glycolipid antigens from damaged cells activate invariant natural killer T (iNKT) cells, which secrete interferon-γ (IFN-γ) and chemokines such as CXCR3, amplifying neutrophil recruitment and promoting inflammation. Elevated numbers of active iNKT cells have been documented during VOCs (65). The complement system also plays a pivotal role in the inflammatory cycle. Heme stimulates ECs to release Weibel-Palade bodies, upregulating surface expression of P-selectin. This molecule interacts with complement component C3b, triggering complement activation. Additionally, heme can promote the release of C5a and C5b-9, which not only exacerbate endothelial injury but also activate inflammatory pathways such as NF-κB (66).

Vaso-occlusion and ischemia-reperfusion injury represent another critical aspect of SCD-associated inflammation. When blood flow is disrupted, tissues become hypoxic, and upon reperfusion, ROS and calcium influx trigger further cellular damage. DAMPs, such as ATP, high-mobility group box 1 (HMGB1), and HSPs, are released, amplifying inflammatory signaling and contributing to a vicious cycle of tissue damage (67). Overall, inflammation is central to the pathophysiology of VOCs in SCD, driving both the onset and progression of these episodes. Studies have consistently shown elevated levels of pro-inflammatory cytokines, particularly IL-8 and IL-17, during active crises, compared to steady-state conditions (68,69).

Coagulation System and Thrombotic Events in SCD

Hemostasis relies on a coordinated response involving blood vessels, platelets, and plasma proteins to maintain blood flow and prevent excessive blood loss. Upon injury, vascular smooth muscle cells (VSMCs) and platelets manage primary hemostasis, while the coagulation factors in both intrinsic and extrinsic pathways act in concert to form stable fibrin clots, initiating secondary hemostasis (70-72). TF, predominantly expressed by perivascular cells such as VSMCs, acts as the key initiator of the extrinsic coagulation pathway (73). While TF expression is essential for preventing blood loss during vascular injury, studies have revealed its increased expression in SCD. This heightened TF expression is evident not only in circulating ECs but also in microparticles (MPs) derived from both ECs and monocytes (74, 75). In addition, TF procoagulant activity is found to be elevated in the whole blood of individuals with SCD (76). The combination of increased TF expression, endothelial injury, and heightened vascular permeability promotes TF exposure to coagulation factors, contributing to clot formation and thrombosis (77, 78). Moreover, sickling and damage to RBCs leads to the translocation of phosphatidylserine (PS) from the inner to the outer leaflet of the RBC membrane,

[DOI: 10.18502/ijpho.v15i3.18922]

generating a negatively charged surface. This negative charge triggers the coagulation cascade through electrostatic interactions with the positively charged γ-carboxyglutamic acid domains present in coagulation factors II, VII, IX, and X (79, 80). Additionally, PS exposure is linked with acquired protein S deficiency in SCD, which accelerates the clearance of protein S from circulation. Since protein S plays a synergistic role in inhibiting coagulation factors V and VIII, its reduced levels further promote hypercoagulability (81, 82). MPs derived from RBCs and platelets activate the intrinsic coagulation pathway through factor XII, whereas those derived from monocytes primarily induce thrombin generation through TF (83). Additionally, VWF levels are elevated in SCD patients, particularly during VOCs, while the activity and levels of ADAMTS13, the enzyme responsible for cleaving VWF, are diminished (84). Several factors contribute to the elevated VWF in SCD, including enhanced production, impaired clearance, and damage to ADAMTS13. The oxidation of ADAMTS13's cleavage site, coupled with free hemoglobin binding to domain A2 of VWF, impairs the enzyme's functionality (85). The persistence of VWF multimers in circulation and their adherence to ECs promote cell adhesion and platelet aggregation, ultimately exacerbating VOCs and thrombotic complications (86, 87).

The level of VWF is also increased in SCD patients, particularly during VOC, while the level and activity of ADAMTS13 decrease during these crises (88, 89). Several factors contribute to the elevated VWF in SCD, including enhanced production, impaired clearance, and damage to ADAMTS13. The oxidation of ADAMTS13's cleavage site, coupled with free Hb binding to domain A2 of VWF, impairs the enzyme's functionality (77, 90, 91). The persistent adherence of VWF multimers to ECs and its increased circulation lead to cell adhesion to ECs and platelet aggregation, resulting in VOC and thrombotic complications (92). Moreover, SCD patients show an increase in antiphospholipid antibodies, particularly IgG-PS, which contribute to both venous and arterial thrombosis by targeting phospholipid-binding proteins (93, 94). Thrombotic activity markers, such as the thrombin-antithrombin complex (TAT) and prothrombin activation fragment (F1.2), are elevated in SCD patients, with TAT levels rising more significantly during VOCs. Fibrinolytic markers, including D-dimers and plasmin-antiplasmin complexes (PAP), also show increased levels, with greater elevations observed in severe VOC episodes (95, 96).

[DOI: 10.18502/ijpho.v15i3.18922]

Table I. Selected Research on the Pathophysiology of VOC in SCD

Topic	Key Findings	Samples	Ref.
Impact of Hb S polymerization factors on clinical outcomes	Clinical severity in SCD predicted by Hb S polymerization, cell heterogeneity, and membrane abnormalities.	46 SS patients	(97)
The role of Hb F in SCA	Elevated Hb F levels inhibit Hb S polymerization, reducing red cell sickling and VOC severity.	272 SS patients	(98)
Erythroid adhesion molecules in SCA	SCA infants show increased expression of adhesion molecules (Lu/BCAM, ICAM-4, LFA-3) on reticulocytes.	54 SCA infants	(99)
Role of oxidative stress, NO, and RBC microparticles in SCA	Oxidative stress promotes eryptosis and RBC microparticle release, which contribute to vascular dysfunction and endothelial inflammation via the TLR4 pathway. Antioxidants and NO improve RBC deformability and reduce eryptosis.	62 SCA HAEC cell line	(100)
Activation of vascular endothelium by stimulated monocytes	Sickle monocytes are activated, producing higher TNF-α and IL-1β levels, leading to endothelial activation and increased adhesion molecule expression.	Sickle mononuclear leukocytes HUVEC cell line MVEC cell line	(101)
Relationship between nocturnal oxyhemoglobin desaturation and VOC complications	Hypoxemia is associated with increased cellular adhesion and activation. Chronic hypoxia contributes to CNS vasculopathy and stroke risk through hypoxia-mediated pathways.	9 SC patients 28 SS patients	(102)
Simultaneous polymerization and adhesion under hypoxia	Young sickle RBCs exhibit unique adhesion dynamics, with polymerized Hb S fiber bundles creating multiple adhesion sites, emphasizing their role in crisis onset.	8 SCA patients	(103)
Neutrophil-Platelet Micro-Emboli Enable VOC	Neutrophils roll, arrest, and capture platelets, forming neutrophil-platelet micro-emboli. Micro-emboli evolve into micro-thrombi, blocking blood flow. Adhesion is mediated by PSGL-1 and Mac-1 on neutrophils binding to P-selectin and GPIbα on platelets.	Unknown SCA patients	(104)
Pro-inflammatory cytokine levels and TGF-β in SCD during VOC and steady state	VOC patients had higher cytokine levels compared to steady-state patients, with IL-8 showing significant increases. Elevated IL-17 and TGF-β levels were noted in steady-state patients versus controls. Hydroxyurea reduced TNF-α, IL-1β, and IL-17 levels	54 SCD patients (39 VOC, 15 steady-state)	(68)
Complement pathway activation during VOC in SCD	Significant elevation in complement activation markers (C3a, C5a, Bb) during VOC compared to the steady state. Complement activation was associated with intravascular hemolysis.	64 SCD patients	(105)
Relationship of coagulation and platelet activation with clinical complications in SCD	Coagulation markers (D-dimer, TAT) were linked to hemolysis indicators and soluble vascular cell adhesion molecule-1. D-dimer was associated with a history of stroke, TAT with retinopathy, and CD40 ligand with pain episodes.	64 SCD patients	(106)

Abbreviations: Hb: Hemoglobin; SCA: Sickle cell anemia; SCD: Sickle cell disease; RBC: Red blood cell; VOC: Vaso-occlusive crisis; HAEC: Human aortic endothelial cells; HUVEC: Human umbilical vein endothelial cells; MVEC: Human dermal microvascular endothelial cells; IL: Interleukin; TNF: Tumor necrosis factor; TAT: Thrombin antithrombin.

[DOI: 10.18502/ijpho.v15i3.18922]

Table II. Summary of Treatment Strategies Related to VOC

Drug	Mechanism	Administration	FDA-approved	Ref.
Hydroxyurea	Increases Hb F production; reduces VOCs and the need for blood transfusions.	Oral	YES	(107)
Butyrate	Stimulates Hb F production.	Oral or infusion	NO	(108)
5-Azacytidine	Acts as a DNA demethylating agent that enhances Hb F production.	Subcutaneous or intravenous	NO	(109)
Decitabine-Tetrahydrouridine	Combination therapy that enhances Hb F through epigenetic modulation.	Intravenous	NO	(110)
L-Glutamine	Reduces oxidative damage to RBCs, lowering the frequency of VOCs.	Oral	YES	(111)
N-Acetylcysteine	Reduces oxidative stress and hemolysis.	Oral or intravenous	NO	(112)
Poloxamer	Inhibits abnormal blood rheology, improves blood flow, and reduces viscosity.	Intravenous	NO	(113)
Prasugrel	Inhibits platelet interactions and thrombosis.	Oral	NO	(114)
Ticagrelor	Acts as a P2Y12 receptor antagonist to reduce platelet activation.	Oral	NO	(115)
Tinzaparin	Inhibits thrombotic activity and prevents blood clot formation.	Subcutaneous	NO	(116)
Inhaled nitric oxide	Reduces endothelial cell dysfunction; acts as a vasodilator to enhance blood flow and reduce cell adhesion.	Inhalation	NO	(117)
Voxelotor	Inhibits Hb S polymerization	Oral	Yes	(118)
Crizanlizumab	Inhibits P-selectin-mediated adhesive interactions and decreases VOCs	Intravenous	Yes	(119)
Arginine therapy	Arginine enhances NO production, improving vasodilation and blood flow in VOC	Oral or intravenous	No	(120)
Exagamglogene autotemcel (exa-cel)	Enhances Hb F production	Intravenous	NO	(121)

Abbreviations: **Hb**; Hemoglobin, **SCD**; Sickle Cell Disease, **RBC**; Red Blood Cell, **VOC**; Vaso-Occlusive Crisis

[DOI: 10.18502/ijpho.v15i3.18922]

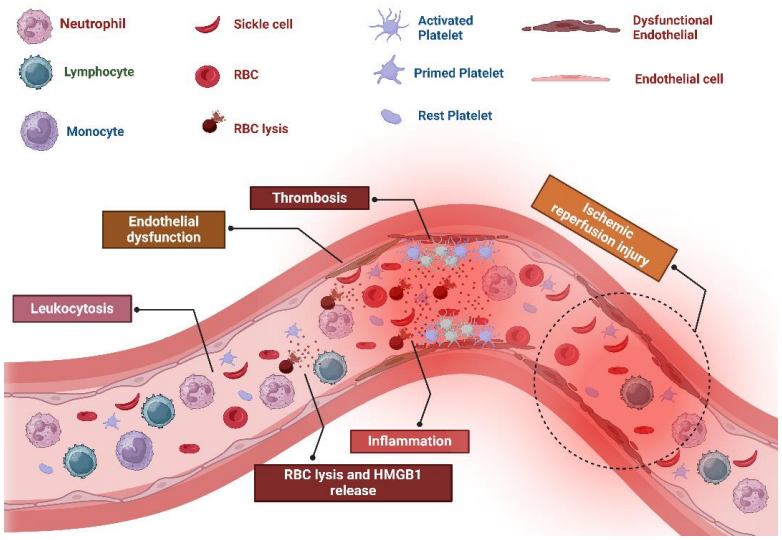

Figure 1. EC Dysfunction as a Central Driver of VOC in SCD.

The figure illustrates the pivotal role of ECs in the initiation and propagation of VOC. Circulating blood components, platelets, WBCs, and sickled RBCs, contribute to a proinflammatory and prothrombotic microenvironment. Activated platelets, in conjunction with leukocytosis, release a cascade of inflammatory cytokines, chemokines, and procoagulant factors. These mediators trigger EC activation and injury, leading to increased vascular permeability, elevated expression of adhesion molecules, and the disruption of endothelial integrity. This dysfunction facilitates enhanced cellular adhesion, thrombin generation, VWF accumulation, and fibrin deposition, ultimately promoting vascular occlusion, impaired blood flow, and recurrent VOC episodes.

RBC: Red blood cell**; VOC:** Vaso-occlusive crisis**; VWF:** von Willebrand factor**; WBC:** White blood cell.

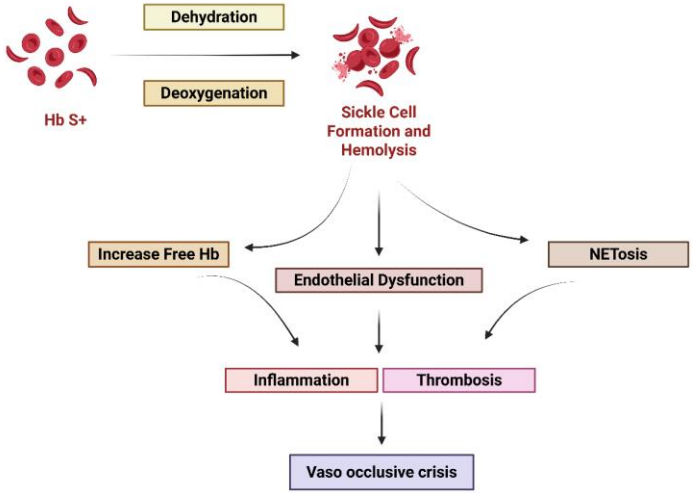

Figure 2. Pathophysiological Cascade of VOC in SCD.

Deoxygenation and dehydration promote Hb S polymerization, leading to red blood cell sickling, increased hemolysis, and reduced deformability. These events initiate endothelial dysfunction and trigger NET formation, collectively amplifying inflammation and thrombotic processes. The resulting immunothrombotic environment exacerbates microvascular occlusion and drives recurrent VOC episodes in SCD.

Hb S: Hemoglobin S; **NET**: Neutrophil extracellular trap; **VOC**: Vaso-occlusive crisis

[DOI: 10.18502/ijpho.v15i3.18922]

Conclusion

VOC in SCD represents a complex, self-amplifying pathological cascade driven by the interconnected processes of hypoxia, inflammation, and coagulation. Hypoxia, initiated by microvascular occlusion and compounded by impaired hemoglobin oxygen delivery, triggers a systemic inflammatory response, mobilizing innate and adaptive immune cells that further damage the endothelium and perpetuate vascular obstruction. Concurrently, hypoxia-induced activation of ECs and platelets facilitates a hypercoagulable state, characterized by TF expression, NETosis, and impaired anticoagulant mechanisms. These thromboinflammatory events not only exacerbate local ischemia but also extend systemically, contributing to multi-organ dysfunction and long-term morbidity in SCD patients.

This triad forms a vicious cycle: hypoxia fuels inflammation; inflammation promotes thrombosis; and coagulation further impairs oxygen delivery, driving the persistence and severity of VOC. Recognizing this interaction is critical not only for understanding the disease mechanism but also for identifying novel therapeutic targets. Interventions aimed at breaking this cycle, whether by modulating inflammasome activation, preserving endothelial integrity, or restoring hemostatic balance, hold promise in alleviating VOC severity and improving outcomes in individuals living with SCD.

Availability of Data and Materials

This is a review study, and it is not an original. Data availability is the responsibility of the corresponding author's.

Ethics Considerations

This article does not contain any studies with human participants or animals performed by any of the authors.

Acknowledgments

We wish thank you of all our coleagues in IRAN University of medical science. During the preparation of this work the author(s) did not use AI.

Author's contribution

Vahid Goodarzi design the study and revise the manuscript. Maedeh Mohammadi, Hakimeh Hadi, Hassan Boustani, Ehsan Kamali Yazdi conduct the write the manuscript.

Funding

The authors stated that no funding was associated with this work.

Conflict of interest

The authors declare that they have no conflict of interest.

References

1. Rees DC, Williams TN, Gladwin MT. Sickle-cell disease. The Lancet 2010; 376(9757):2018-2031.
2. Maddah R, Etemad S, Amiri BS, Ghaderi H, Zarei H, Faghihkhorasani F, et al. Evaluation of genes and molecular pathways involved in pathogenesis of sickle cell anemia: a bioinformatics analysis and future perspective. Iran J Public Health 2024; 53(6): 1404-1415.
3. Jang T, Poplawska M, Cimpeanu E, Mo G, Dutta D, Lim SH. Vaso-occlusive crisis in sickle cell disease: a vicious cycle of secondary events. J Transl Med 2021; 19(1):397-402.
4. Weaver SB, Rungkitwattanakul D, Singh D. Contemporary management and prevention of vaso-occlusive crises (vocs)

[DOI: 10.18502/ijpho.v15i3.18922]

in adults with sickle cell disease. J Pharm Pract 2023; 36(1):139-148.
5. Noronha SA. Cardiac causes of hypoxia in sickle cell disease. Prog Pediat Cardiol 2020; 56:101192-101198.
6. Ahmadipour M, Naghibzadeh-Tahami A, Mirzie K, Maleki E. Vitamin D and calcium deficiency and its relationship with cardiac function in patients with beta thalassemia. J Kerman Univ Med Sci 2024; 31(1):29-34.
7. Wang Q, Zennadi R. The role of RBC oxidative stress in sickle cell disease: from the molecular basis to pathologic implications. Antioxidants (Basel) 2021; 10(10), 35-45.
8. Conran N, Embury SH. Sickle cell vaso-occlusion: The dialectic between red cells and white cells. Exp Biol Med (Maywood) 2021; 246(12):1458-1472.
9. Amin A, Mohajerian A, Ghalehnoo SR, Mohamadinia M, Ahadi S, Sohbatzadeh T, et al. Potential player of platelet in the pathogenesis of cardiotoxicity: Molecular insight and future perspective. Cardiovasc Toxicol 2024; 24(12):1381-1394.
10. Yaykasli KO, Schauer C, Muñoz LE, Mahajan A, Knopf J, Schett G, et al. Neutrophil extracellular trap-driven occlusive diseases. Cells 2021; 10(9): 2208-2211.
11. Ballas SK. Pathophysiology and principles of management of the many faces of the acute vaso-occlusive crisis in patients with sickle cell disease. Eur J Haematol 2015; 95(2):113-123.
12. Ballas SK, Mohandas N. Pathophysiology of vaso-occlusion. Hematol Oncol Clin North Am 1996; 10(6):1221-1239.
13. Yale SH, Nagib N, Guthrie T. Approach to the vaso-occlusive crisis in adults with sickle cell disease. Am Fam Physician 2000; 61(5):1349-1356.
14. Rastegar-Kashkouli A, Yousefi P, Jafari M, Taravati AM, Shahrokh SG, Rastegar D. Sickle cell nephropathy; exploring epidemiology, diagnostic challenges, and precision medicine advances. J Prev Epidemiol 2024; 9(2):e35240-e35240.
15. Zeinali M, Babadi AJ, Barani L. Brown-Séquard syndrome caused by multiple knife trauma gunshot with late debridement: Two cases report and literature review. Int J Surg Case Rep 2023; 105:108068-108075.
16. Emami M, Shafie D, Vakhshoori M, Eghbali-Babadi M, Ahmadipour E, Khosravi A. Evaluation of the efficiency of indirect blood pressure measurement methods in comparison to intra-arterial reading among iranian individuals. Int Cardiovasc Res J 2020; 14(3):e103852-e103852.
17. Osunkwo I, Manwani D, Kanter J. Current and novel therapies for the prevention of vaso-occlusive crisis in sickle cell disease. Ther Adv Hematol 2020; 11: 2040620720955000-2040620720955000.
18. Vekilov PG. Sickle-cell haemoglobin polymerization: is it the primary pathogenic event of sickle-cell anaemia? Br J Haematol 2007; 139(2):173-184.
19. Allali S, Rignault-Bricard R, de Montalembert M, Taylor M, Bouceba T, Hermine O, et al. HbS promotes TLR4-mediated monocyte activation and proinflammatory cytokine production in sickle cell disease. Blood 2022 3; 140(18):1972-1982.
20. Shafique F, Ali S, Almansouri T, Van Eeden F, Shafi N, Khalid M, et al. Thalassemia, a human blood disorder. Braz J Biol 2021 3; 83:e246062- e246062.
21. Eaton WA, Bunn HF. Treating sickle cell disease by targeting HbS polymerization. Blood 2017; 129(20):2719-2726.
22. Aghaei HN, Azimi P, Shahzadi S, Azhari S, Mohammadi HR, Alizadeh P, et al. Outcome measures of functionality, social interaction, and pain in patients with cervical spondylotic myelopathy: a validation study for the iranian version of the copenhagen neck functional disability scale. Asian Spine J 2015; 9(6):901-905.

[DOI: 10.18502/ijpho.v15i3.18922]

23. Akinsheye I, Alsultan A, Solovieff N, Ngo D, Baldwin CT, Sebastiani P, et al. Fetal hemoglobin in sickle cell anemia. Blood 2011; 118(1):19-27.
24. Elendu C, Amaechi DC, Alakwe-Ojimba CE, Elendu TC, Elendu RC, Ayabazu CP, et al. Understanding sickle cell disease: Causes, symptoms, and treatment options. Medicine (Baltimore) 2023; 102(38):e35237- e35237.
25. Sumpio BE, Riley JT, Dardik A. Cells in focus: endothelial cell. Int J Biochem Cell Biol 2002; 34(12):1508-1512.
26. Hebbel RP, Vercellotti GM. The endothelial biology of sickle cell disease. J Lab Clin Med 1997; 129(3):288-293.
27. Inusa BPD, Hsu LL, Kohli N, Patel A, Ominu-Evbota K, Anie KA, et al. Sickle cell disease-genetics, pathophysiology, clinical presentation and treatment. Int J Neonatal Screen 2019; 5(2):20-27.
28. White J, Lancelot M, Gao X, McGraw BJ, Tabb C, 2nd, Hines P. Cross-sectional analysis of adhesion in individuals with sickle cell disease using a standardized whole blood adhesion bioassay to VCAM-1. Blood Cells Mol Dis 2020; 81:102397-102490.
29. Brandow AM, Liem RI. Advances in the diagnosis and treatment of sickle cell disease. J Hematol Oncol 2022; 15(1):20.
30. Piccin A, Murphy C, Eakins E, Rondinelli MB, Daves M, Vecchiato C, et al. Insight into the complex pathophysiology of sickle cell anaemia and possible treatment. Eur J Haematol 2019; 102(4):319-330.
31. Kazemi N, Bordbar A, Bavarsad SS, Ghasemi P, Bakhshi M, Rezaeeyan H. Molecular insights into the relationship between platelet activation and endothelial dysfunction: Molecular approaches and clinical practice. Mol Biotechnol 2024; 66(5):932-947.
32. Shet AS, Lizarralde-Iragorri MA, Naik RP. The molecular basis for the prothrombotic state in sickle cell disease. Haematologica 2020; 105(10):2368-2372.
33. Hebbel RP, Vercellotti GM. Multiple inducers of endothelial NOS (eNOS) dysfunction in sickle cell disease. Am J Hematol 2021; 96(11):1505-1517.
34. Ataga KI, Kutlar A, Kanter J, Liles D, Cancado R, Friedrisch J, et al. Crizanlizumab for the prevention of pain crises in sickle cell disease. N Engl J Med 2017; 376(5):429-439.
35. Dampier CD, Telen MJ, Wun T, Brown RC, Desai P, El Rassi F, et al. A randomized clinical trial of the efficacy and safety of rivipansel for sickle cell vaso-occlusive crisis. Blood 2023; 141(2):168-179.
36. Homi J, Levee L, Higgs D, Thomas P, Serjeant G. Pulse oximetry in a cohort study of sickle cell disease. Clin Lab Haematol 1997; 19(1):17-22.
37. Di Liberto G, Kiger L, Marden MC, Boyer L, Poitrine FC, Conti M, et al. Dense red blood cell and oxygen desaturation in sickle-cell disease. Am J Hematol 2016; 91(10):1008-1013.
38. Abdu A, Gómez-Márquez J, Aldrich TK. The oxygen affinity of sickle hemoglobin. Respir Physiol Neurobiol 2008; 161(1):92-94.
39. Sears DA, Udden MM, Thomas LJ. Carboxyhemoglobin levels in patients with sickle-cell anemia: relationship to hemolytic and vasoocclusive severity. Am J Med Sci 2001; 322(6):345-348.
40. Ajudani R, Alizadeh P, Zeinali M, Jafari Marandi H. Determining the optimal morphometry of pedicle screws in lumbar and thoracic spine in the iranian population. J Iran Med Counc 2024; 7(4):776-783.
41. Caboot JB, Allen JL. Hypoxemia in sickle cell disease: significance and management. Paediatr Respir Rev 2014; 15(1):17-23.
42. Moradi G, Etehadi SS, Mousavi SH, Mohammadi R. Evaluating the cavitary lung lesions on CT scan of COVID-19 patients: A retrospective study.

[DOI: 10.18502/ijpho.v15i3.18922]

J Community Hosp Intern Med Perspect 2024; 14(6):23-29.
43. Schechter AN, Gladwin MT. Hemoglobin and the paracrine and endocrine functions of nitric oxide. N Engl J Med 2003; 348(15):1483-1485.
44. Sullivan KJ, Goodwin SR, Evangelist J, Moore RD, Mehta P. Nitric oxide successfully used to treat acute chest syndrome of sickle cell disease in a young adolescent. Crit Care Med 1999; 27(11):2563-2568.
45. Head CA, Brugnara C, Martinez-Ruiz R, Kacmarek RM, Bridges KR, Kuter D, et al. Low concentrations of nitric oxide increase oxygen affinity of sickle erythrocytes in vitro and in vivo. J Clin Invest 1997; 100(5):1193-1198.
46. Martinez-Ruiz R, Montero-Huerta P, Hromi J, Head CA. Inhaled nitric oxide improves survival rates during hypoxia in a sickle cell (SAD) mouse model. Anesthesiology 2001; 94(6):1113-1118.
47. Gladwin MT, Kato GJ, Weiner D, Onyekwere OC, Dampier C, Hsu L, et al. Nitric oxide for inhalation in the acute treatment of sickle cell pain crisis: a randomized controlled trial. Jama 2011; 305(9):893-902.
48. Sun K, Xia Y. New insights into sickle cell disease: a disease of hypoxia. Curr Opin Hematol 2013; 20(3):215-221.
49. Rad HF, Karimi MS, Behfar M, Nazari A, Hassani N, Hasanvand A, et al. LncRNA Linc00261 sponge's miR-33-regulated PI3-kinase in autism disorder. Gene Reports 2025; 38:102115-102120.
50. Tan F, Ghosh S, Mosunjac M, Manci E, Ofori-Acquah SF. Diametric effects of hypoxia on pathophysiology of sickle cell disease in a murine model. Exp Biol Med (Maywood) 2016; 241(7):766-771.
51. Hebbel RP, Belcher JD, Vercellotti GM. The multifaceted role of ischemia/reperfusion in sickle cell anemia. J Clin Invest 2020; 130(3):1062-1072.
52. Zipursky A, Robieux IC, Brown EJ, Shaw D, O'Brodovich H, Kellner JD, et al. Oxygen therapy in sickle cell disease. Am J Pediatr Hematol Oncol 1992; 14(3):222-228.
53. Reiter CD, Wang X, Tanus-Santos JE, Hogg N, Cannon RO, Schechter AN, et al. Cell-free hemoglobin limits nitric oxide bioavailability in sickle-cell disease. Nat Med 2002; 8(12):1383-1389.
54. Antwi-Boasiako C, Dankwah G, Aryee R, Hayfron-Benjamin C, Donkor E, Campbell A. Oxidative profile of patients with sickle cell disease. Med Sci (Basel) 2019; 7(2):17-22.
55. Hebbel RP, Morgan WT, Eaton JW, Hedlund BE. Accelerated autoxidation and heme loss due to instability of sickle hemoglobin. Proc Natl Acad Sci 1988; 85(1):237-241.
56. Schmidt HM, Wood KC, Lewis SE, Hahn SA, Williams XM, McMahon B, et al. Xanthine oxidase drives hemolysis and vascular malfunction in sickle cell disease. Arterioscler Thromb Vasc Biol 2021; 41(2):769-782.
57. Marçal LE, Dias-da-Motta PM, Rehder J, Mamoni RL, Blotta MHS, Whitney CB, et al. Up-regulation of NADPH oxidase components and increased production of interferon-gamma by leukocytes from sickle cell disease patients. Am J Hematol 2008; 83(1):41-45.
58. Aslan M, Ryan TM, Adler B, Townes TM, Parks DA, Thompson JA, et al. Oxygen radical inhibition of nitric oxide-dependent vascular function in sickle cell disease. Proc Natl Acad Sci U S A 2001; 98(26):15215-15220.
59. Schimmel M, Nur E, Biemond BJ, van Mierlo GJ, Solati S, Brandjes DP, et al. Nucleosomes and neutrophil activation in sickle cell disease painful crisis. Haematologica 2013; 98(11):1797-1799.
60. Ansari J, Gavins FN. Ischemia-reperfusion injury in sickle cell disease: from basics to therapeutics. Am J Pathol 2019; 189(4):706-718.
61. Nazari SH, Azimi MR, Valinezhad S. White cheese physicochemical properties made of IR pasteurized milk ripened in KCL solution. CABA 2017:14-19.

[DOI: 10.18502/ijpho.v15i3.18922]

62. de Zoete MR, Palm NW, Zhu S, Flavell RA. Inflammasomes. Cold Spring Harb Perspect Biol 2014; 6(12):a016287-a016287.
63. Chen G, Zhang D, Fuchs TA, Manwani D, Wagner DD, Frenette PS. Heme-induced neutrophil extracellular traps contribute to the pathogenesis of sickle cell disease. Blood 2014; 123(24):3818-3827.
64. Proenca-Ferreira R, Brugnerotto AF, Garrido VT, Dominical VM, Vital DM, Ribeiro MdFR, et al. Endothelial activation by platelets from sickle cell anemia patients. PloS one 2014; 9(2):e89012- e89012.
65. Field JJ, Lin G, Okam MM, Majerus E, Keefer J, Onyekwere O, et al. Sickle cell vaso-occlusion causes activation of iNKT cells that is decreased by the adenosine A2A receptor agonist regadenoson. Blood 2013; 121(17):3329-3334.
66. Tampaki A, Gavriilaki E, Varelas C, Anagnostopoulos A, Vlachaki E. Complement in sickle cell disease and targeted therapy: I know one thing, that I know nothing. Blood Rev 2021; 48:100805-100805.
67. Conran N, Belcher JD. Inflammation in sickle cell disease. Clin Hemorheol Microcirc 2018; 68(2-3):263-299.
68. Keikhaei B, Mohseni AR, Norouzirad R, Alinejadi M, Ghanbari S, Shiravi F, et al. Altered levels of pro-inflammatory cytokines in sickle cell disease patients during vaso-occlusive crises and the steady state condition. Eur Cytokine Netw 2013; 24(1):45-52.
69. Tran H, Gupta M, Gupta K. Targeting novel mechanisms of pain in sickle cell disease. Blood 2017; 130(22):2377-2385.
70. Versteeg HH, Heemskerk JW, Levi M, Reitsma PH. New fundamentals in hemostasis. Physiol Rev 2013; 93(1):327-358.
71. Wong KC. CORR insights®: comparison of two modern survival prediction tools, SORG-MLA and METSSS, in patients with symptomatic long-bone metastases who underwent local treatment with surgery followed by radiotherapy and with radiotherapy alone. Clin Orthop Relat Res 2024 1; 482(12):2209-2211.
72. Peng D, Bai W, Zhang C, Chang X, Ma P, Wang XL. X-ray irradiation effectively inactivated lymphocytes in transfusion in vivo monitored by the bioluminescence transfusion-associated graft-versus-host disease model. Vox Sang 2024; 119(3):181-192.
73. Østerud B, Bjørklid E. Sources of tissue factor. Semin Thromb Hemost 2006 ; 32(1):11-23.
74. Solovey A, Gui L, Key NS, Hebbel RP. Tissue factor expression by endothelial cells in sickle cell anemia. J Clin Invest 1998; 101(9):1899-1904.
75. Shet AS, Aras O, Gupta K, Hass MJ, Rausch DJ, Saba N, et al. Sickle blood contains tissue factor–positive microparticles derived from endothelial cells and monocytes. Blood 2003; 102(7):2678-2683.
76. Key NS, Slungaard A, Dandelet L, Nelson SC, Moertel C, Styles LA, et al. Whole blood tissue factor procoagulant activity is elevated in patients with sickle cell disease. Blood 1998; 91(11):4216-4223.
77. Sparkenbaugh E, Pawlinski R. Interplay between coagulation and vascular inflammation in sickle cell disease. Br J Haematol 2013; 162(1):3-14.
78. Mirfakhraee H, Larijani SS, Zandieh Z, Tizmaghz A, Olamaeian F, Tayebi A, et al. Renal vein thrombosis in a recent COVID-19 patient; a case report. J Renal Inj Prev 2022; 12(1):e31997-e31997.
79. Li H, Qiang Y, Li X, Brugnara C, Buffet PA, Dao M, et al. Biomechanics of phagocytosis of red blood cells by macrophages in the human spleen. Proc

[DOI: 10.18502/ijpho.v15i3.18922]

Natl Acad Sci 2024; 121(44):e2414437121- e2414437121.
80. Setty BY, Kulkarni S, Rao AK, Stuart MJ. Fetal hemoglobin in sickle cell disease: relationship to erythrocyte phosphatidylserine exposure and coagulation activation. Blood 2000; 96(3):1119-1124.
81. Whelihan M, Lim M, Mooberry M, Piegore M, Ilich A, Wogu A, et al. Thrombin generation and cell-dependent hypercoagulability in sickle cell disease. J Thromb Haemost 2016; 14(10):1941-1952.
82. Suleiman L, Négrier C, Boukerche H. Protein S: A multifunctional anticoagulant vitamin K-dependent protein at the crossroads of coagulation, inflammation, angiogenesis, and cancer. Crit Rev Oncol Hematol 2013; 88(3):637-654.
83. Van Der Meijden P, Van Schilfgaarde M, Van Oerle R, Renné T, Ten Cate H, Spronk H. Platelet-and erythrocyte-derived microparticles trigger thrombin generation via factor XIIa. J Thromb Haemost 2012; 10(7):1355-1362.
84. Wun T, Paglieroni T, Rangaswami A, Hill Franklin P, Welborn J, Cheung A, et al. Platelet activation in patients with sickle cell disease. Br J Haematol 1998; 100(4):741-749.
85. Mehta P, Mehta J. Abnormalities of platelet aggregation in sickle cell disease. J Pediatr 1980; 96(2):209-213.
86. Browne PV, Mosher DF, Steinberg MH, Hebbel RP. Disturbance of plasma and platelet thrombospondin levels in sickle cell disease. Am J Hematol 1996; 51(4):296-301.
87. Ataga KI, Orringer EP. Hypercoagulability in sickle cell disease: a curious paradox. Am J Med 2003; 115(9):721-728.
88. Krishnan S, Siegel J, Pullen Jr G, Hevelow M, Dampier C, Stuart M. Increased von Willebrand factor antigen and high molecular weight multimers in sickle cell disease associated with nocturnal hypoxemia. Thromb Res 2008; 122(4):455-458.
89. Sins JW, Schimmel M, Luken BM, Nur E, Zeerleder SS, van Tuijn CF, et al. Dynamics of von Willebrand factor reactivity in sickle cell disease during vaso-occlusive crisis and steady state. J Thromb Haemost 2017; 15(7):1392-1402.
90. Zhou Z, Han H, Cruz MA, López JA, Dong J-F, Guchhait P. Haemoglobin blocks von Willebrand factor proteolysis by ADAMTS-13: a mechanism associated with sickle cell disease. Thromb Haemost 2009; 101(06):1070-1077.
91. Jalili C, Darakhshan S, Azimi M. Harmine mitigates liver injury induced by mercuric chloride via the inhibition of oxidative stress. Res J Pharmacogn 2021; 8(3):13-23.
92. Wick T, Moake J, Udden M, Eskin S, Sears D, McIntire L. Unusually large von Willebrand factor multimers increase adhesion of sickle erythrocytes to human endothelial cells under controlled flow. J Clin Invest 1987; 80(3):905-910.
93. Westerman MP, Green D, Gilman-Sachs A, Beaman K, Freels S, Boggio L, et al. Antiphospholipid antibodies, proteins C and S, and coagulation changes in sickle cell disease. J Lab Clin Med 1999; 134(4):352-362.
94. Triplett DA. Antiphospholipid antibodies. Arch Pathol Lab Med 2002; 126(11):1424-1429.
95. Tomer A, Harker LA, Kasey S, Eckman JR. Thrombogenesis in sickle cell disease. J Lab Clin Med 2001; 137(6):398-407.
96. Salesi M, Ansari N. Antiphospholipid antibody nephropathy; an updated mini-review. J Nephropathol 2025; 14(1):e21529-e21529.
97. Poillon WN, Kim BC, Castro O. Intracellular hemoglobin S polymerization and the clinical severity of sickle cell anemia. Blood 1998; 91(5):1777-1783.
98. Demirci S, Leonard A, Essawi K, Tisdale JF. CRISPR-Cas9 to induce fetal hemoglobin for the treatment of sickle cell

[DOI: 10.18502/ijpho.v15i3.18922]

disease. Mol Ther Methods Clin Dev 2021 1; 23:276-285.
99. Brousse V, Colin Y, Pereira C, Arnaud C, Odièvre MH, Boutemy A, et al. Erythroid adhesion molecules in sickle cell anaemia infants: insights into early pathophysiology. EBioMedicine 2015; 2(2):154-157.
100. Nader E, Romana M, Guillot N, Fort R, Stauffer E, Lemonne N, et al. Association between nitric oxide, oxidative stress, eryptosis, red blood cell microparticles, and vascular function in sickle cell anemia. Front Immunol 2020; 11:551441-551445.
101. Belcher JD, Marker PH, Weber JP, Hebbel RP, Vercellotti GM. Activated monocytes in sickle cell disease: potential role in the activation of vascular endothelium and vaso-occlusion. Blood 2000; 96(7):2451-2459.
102. Setty BY, Stuart MJ, Dampier C, Brodecki D, Allen JL. Hypoxaemia in sickle cell disease: biomarker modulation and relevance to pathophysiology. The Lancet 2003; 362(9394):1450-1455.
103. Papageorgiou DP, Abidi SZ, Chang H-Y, Li X, Kato GJ, Karniadakis GE, et al. Simultaneous polymerization and adhesion under hypoxia in sickle cell disease. Proc Natl Acad Sci U S A 2018; 115(38):9473-9478.
104. Sundd P, Jimenez M, Novelli EM, Gladwin MT. Neutrophil-platelet micro-emboli enable vaso-occlusion in sickle cell disease. Blood 2014; 124(21):2695.
105. Yoo JJ, Graciaa SH, Jones JA, Zuo Z, Arthur CM, Leong T, et al. Complement activation during vaso-occlusive pain crisis in pediatric sickle cell disease. Blood 2021; 138:858.
106. Ataga KI, Brittain JE, Desai P, May R, Jones S, Delaney J, et al. Association of coagulation activation with clinical complications in sickle cell disease. PloS one 2012; 7(1):e29786-e29786.
107. Steinberg MH, McCarthy WF, Castro O, Ballas SK, Armstrong FD, Smith W, et al. The risks and benefits of long-term use of hydroxyurea in sickle cell anemia: a 17.5 year follow-up. Am J Hematol 2010; 85(6):403-408.
108. Atweh GF, Sutton M, Nassif I, Boosalis V, Dover GJ, Wallenstein S, et al. Sustained induction of fetal hemoglobin by pulse butyrate therapy in sickle cell disease. Blood 1999; 93(6):1790-1797.
109. Ma YN, Zhang X, Yu HC, Zhang JW. CTD small phosphatase like 2 (CTDSPL2) can increase ε- and γ-globin gene expression in K562 cells and CD34+ cells derived from umbilical cord blood. BMC Cell Biol 2010; 11:75-79.
110. Andemariam B, Højlund Nielsen P, Zillmer R, Derving Karsbøl J. ASCENT1: a phase 2 trial to evaluate the efficacy and Safety of oral decitabine-tetrahydrouridine (NDec) in patients with sickle cell disease. Blood 2022; 140(Supplement 1):5420-5421.
111. Niihara Y, Miller ST, Kanter J, Lanzkron S, Smith WR, Hsu LL, et al. A phase 3 trial of l-glutamine in sickle cell disease. N Engl J Med 2018; 379(3):226-235.
112. Nur E, Brandjes DP, Teerlink T, Otten H-M, Oude Elferink RP, Muskiet F, et al. N-acetylcysteine reduces oxidative stress in sickle cell patients. Ann Hematol 2012; 91:1097-1105.
113. Orringer EP, Casella JF, Ataga KI, Koshy M, Adams-Graves P, Luchtman-Jones L, et al. Purified poloxamer 188 for treatment of acute vaso-occlusive crisis of sickle cell disease: a randomized controlled trial. Jama 2001; 286(17):2099-2106.
114. Jakubowski JA, Zhou C, Jurcevic S, Winters KJ, Lachno DR, Frelinger III AL, et al. A phase 1 study of prasugrel in patients with sickle cell disease: effects on biomarkers of platelet activation and coagulation. Thromb Res 2014; 133(2):190-195.
115. Ribeiro-Filho J, Yahouédéhou SCMA, Pitanga TN, Santana SS, Adorno EV, Barbosa CG, et al. An evaluation of

[DOI: 10.18502/ijpho.v15i3.18922]

ticagrelor for the treatment of sickle cell anemia. Expert Rev Hematol 2020; 13(10):1047-1055.
116. Qari MH, Aljaouni SK, Alardawi MS, Fatani H, Alsayes FM, Zografos P, et al. Reduction of painful vaso-occlusive crisis of sickle cell anaemia by tinzaparin in a double-blind randomized trial. Thromb Haemost 2007; 98(08):392-396.
117. Weiner DL, Hibberd PL, Betit P, Cooper AB, Botelho CA, Brugnara C. Preliminary assessment of inhaled nitric oxide for acute vaso-occlusive crisis in pediatric patients with sickle cell disease. Jama 2003; 289(9):1136-1142.
118. Howard J, Ataga KI, Brown RC, Achebe M, Nduba V, El-Beshlawy A, et al. Voxelotor in adolescents and adults with sickle cell disease (HOPE): long-term follow-up results of an international, randomised, double-blind, placebo-controlled, phase 3 trial. Lancet Haematol 2021; 8(5):e323-e333.
119. Abboud MR, Cançado RD, De Montalembert M, Smith WR, Rimawi H, Voskaridou E, et al. Crizanlizumab with or without hydroxyurea in patients with sickle cell disease (STAND): primary analyses from a placebo-controlled, randomised, double-blind, phase 3 trial. Lancet Haematol 2025; 12(4):e248-e257.
120. Rees CA, Brousseau DC, Cohen DM, Villella A, Dampier C, Brown K, et al. Sickle cell disease treatment with arginine therapy (STArT): study protocol for a phase 3 randomized controlled trial. Trials 2023; 24(1):538-542.
121. Frangoul H, Locatelli F, Sharma A, Bhatia M, Mapara M, Molinari L, et al. Exagamglogene autotemcel for severe sickle cell disease. N Engl J Med 2024; 390(18):1649-1662.

[DOI: 10.18502/ijpho.v15i3.18922]