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Differences in the clinical and genotypic presentation of sickle cell disease around the world

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Summary

Sickle cell disease (SCD), caused by a mutation in the β -globin gene *HBB*, is widely distributed in malaria endemic regions. Cardiopulmonary complications are major causes of morbidity and mortality. Hemoglobin SS (Hb SS) represents a large proportion of SCD in the Americas, United Kingdom, and certain regions of Africa while higher proportions of hemoglobin SC are observed in Burkina Faso and hemoglobin S β -thalassemia in Greece and India. Coinheritance of α -thalassemia and persistence of hemoglobin F production are observed in highest frequency in certain regions of India and the Middle East. As confirmed in the PUSH and Walk-PHaSST studies, Hb SS, absence of co-inheriting alpha-thalassemia, and low hemoglobin F levels tend to be associated with more hemolysis, lower hemoglobin oxygen saturations, greater proportions of elevated tricuspid regurgitant jet velocity and brain natriuretic peptide, and increased left

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ventricular mass index. Identification of additional genetic modifiers will improve prediction of cardiopulmonary complications in SCD.

Keywords

Sickle Cell Disease; Echocardiogram; Pulmonary Function Test; Hemoglobin F; Alpha thalassemia

Introduction

Sickle cell disease (SCD) is the consequence of homozygosity for a single amino acid change in the β -globin chain that results in structurally abnormal hemoglobin S, or by compound heterozygosity for hemoglobin S and another β -globin chain abnormality, typically hemoglobin C or β -thalassemia. In addition, α -thalassemia is a modifier of the clinical manifestations of SCD. The sickle-cell gene, β -thalassemia and α -thalassemia are distributed widely throughout sub-Saharan Africa the Middle East and parts of the Indian subcontinent, all areas of high prevalence of malaria historically. This striking overlap in the historical distribution of malaria and hemoglobin S, β -thalassemia and α -thalassemia provides evidence that protection from malaria mortality is what provided a heterozygote advantage for these mutations to become prevalent in certain populations. Carrier rates for hemoglobin S range from 5% to 40% or more of the population in these areas (1).

Sickle cell disease is among the most common monogenetic diseases worldwide (2). It is estimated that 312,000 people with hemoglobin SS (Hb SS) are born each year throughout the world, with the majority of these births (236,000) in sub-Saharan Africa (3). Based on the World Health Organization published global prevalence map of SCD and other data (<http://www.who.int/genomics/public/Maphaemoglobin.pdf>), about 20–25 million individuals world-wide have homozygous SCD; 12–15 million in sub-Saharan Africa, 5–10 million in India and about 3 million distributed in other parts of the world. Migration patterns have led to the distribution of the sickle cell genes into regions that are not endemic for malaria. About 8% of African Americans carry the sickle gene (4, 5) and the expected incidence of SCD at birth is 1 in 625 (6, 7). Approximately 100,000 people with SCD live in the United States (8).

Hemoglobin S polymerizes upon deoxygenation and this causes erythrocyte rigidity, sickling and early destruction. Vaso-occlusion and hemolysis related to these rigid and/or sickled cells lead to disease manifestations, including hemolytic anemia, severe pain episodes from bone marrow ischemia, central nervous system strokes, the acute chest syndrome, pulmonary hypertension, left-sided heart disease, bacteremia, leg ulcers, growth failure, priapism and damage to the spleen, kidneys, liver and bones (9). The disease severity varies considerably among patients with SCD and even among those with Hb SS. While some patients have severe complications and die before the third decade, others may remain largely asymptomatic. For example, in an investigation of pain involving 3578 patients as a part of the Cooperative Study of Sickle Cell Disease (CSSCD), 5.2 % had 3–10 painful episodes requiring medical care per year while 39% had none (10).

Prominent recognized factors that influence the severity of SCD include the genotype, for example Hb SS versus hemoglobin SC (Hb SC) versus hemoglobin S β -thalassemia (Hb S β -thalassemia), the coexistent presence of α -thalassemia, and the level of hemoglobin F (Hb F). We will consider the geographic distribution of these factors and how they influence the pulmonary and cardiac complications of SCD. This paper does not represent a formal meta-

analysis or a comprehensive review of the literature. Rather it provides selected examples of geographic variation and prevalences.

Distribution of sickle cell disease genotypes in patients of predominantly African origin in the Americas and the UK

The predominant genotypes that give rise to SCD include Hb SS, Hb SC, Hb S β^+ -thalassemia and Hb S β^0 -thalassemia. Other rare forms include hemoglobin SD and hemoglobin SE. Three recent large multicenter cohorts provide the distribution of these genotypes among SCD patients of predominantly African origin in the Americas and the UK (Table 1). In a cross-sectional study conducted at three centers in Brazil, 773 children and adolescents 2–16 years of age were screened with transcranial Doppler ultrasound (11). In the Pulmonary Hypertension and the Hypoxic Response in SCD (PUSH) study conducted at four tertiary medical centers in the US, 720 individuals 3 to 20 years of age were screened with echocardiography as outpatients at steady-state (12). In the Walk-PHaSST (treatment of Pulmonary Hypertension and Sickle cell disease with Sildenafil Therapy) study individuals 12 years with SCD were screened with echocardiography as outpatients at steady-state at nine US Centers and one UK Center, with 89.3% of the patients being over 20 years of age (13). In these three cohorts, the proportion of Hb SS genotype ranged from 74–76%, of Hb SC from 18–24%, and of Hb S β -thalassemia from 1–6%.

Geographic variability of distribution of sickle cell disease genotypes outside the Americas and UK

Given the fairly uniform distribution of sickle genotypes in the Americas and the UK, it is striking how different the distributions are in specific geographic areas where malaria is endemic (Table 2). Among three university hospital-based cohorts from Nigeria and Senegal, the proportion of Hb SS genotype ranged from 88–96%, of Hb SC from 4–12%, and Hb S β -thalassemia <1% (14–16). In contrast, among 153 SCD individuals identified through screening programs in villages or schools in the area of Ouagadougou, Burkina Faso, 92% had the Hb SC genotype and 8% had the Hb SS (17). The genotype distribution among 387 SCD patients diagnosed by performing hemoglobin electrophoresis in an urban hospital in Ouagadougou was 50% Hb SC and 50% Hb SS (18), likely representing an increased likelihood for Hb SS patients to present to the hospital with problems requiring diagnosis and treatment. Further variation is shown in cohorts from Greece (19) and Chennai, India (20), where the predominant genotype is Hb S β -thalassemia but Hb SS comprises 19–36% of the cohorts, and in a cohort from Andhra Pradesh, India (21), where the predominant genotype is Hb SS but Hb S β -thalassemia makes up 31% of the cohort.

Geographic variability of α -thalassemia in sickle cell disease

The α -thalassemias are frequently encountered in malaria endemic regions of Southeast Asia, Africa, India, and the Middle East. The α -globin genes are duplicated on each copy of chromosome 16 for a total of four α -globin genes ($\alpha\alpha/\alpha\alpha$). Deletions (e.g. $-\alpha^{3.7}$ and $-\alpha^{4.2}$) or gene mutations (e.g. $\alpha^{\text{Constant Spring}}$ and α^{TSaudi}) result in decreased production of α -globin chains. In Southeast Asia, the pattern of *cis*-deletions ($--/\alpha\alpha$) are more common in α -thalassemia with a prevalence of 4 – 14% and some regions reporting prevalences near 80% (22, 23). The hemoglobin Constant Spring mutation is also frequently observed in the Southeast Asia region with frequencies of 1 – 6% (24). In contrast to the Southeast Asia region pattern of *cis*-deletions, *trans*-deletions ($-\alpha/-\alpha$) are more frequently observed in people from African, Indian, and Middle Eastern regions. The ($-\alpha$) chromosome frequency varies through regions of Africa with the highest rates observed in central Africa.

Frequencies of the $(-\alpha)$ chromosome range from 0.14 – 0.36 in Hb AA populations and 0.24 – 0.45 in Hb SS populations of central Africa (25–28) while lower frequencies between 0.05 – 0.11 are observed in Hb AA and SS populations of northern and southern Africa (29–32). In regions of India, the frequency of the $(-\alpha)$ chromosome is 0.10 – 0.22 in the general population, 0.24 – 0.62 in Hb SS populations, and 0.97 in certain tribal populations with Hb SS (33–35). High rates of α -thalassemia are also observed in the Middle East with estimates of up to 50% of the population carrying deletional ($-\alpha^{3.7}$ and $-\alpha^{4.2}$) or nondeletional ($\alpha^{\text{Constant Spring}}$ and α^{TSaudi}) forms of α -thalassemia (36). In cohorts of Hb SS populations from the Middle East, frequencies of the $(-\alpha)$ chromosome are 0.39 – 0.48 (37, 38). Migration patterns have led to the distribution of α -thalassemia deletion and non-deletion mutations in non-malaria endemic regions. In the general population of African Americans, the frequency of the $(-\alpha)$ chromosome is estimated at 0.16 and is similar to that observed in African Brazilians (39, 40). Among Hb SS patients from North and South America, the $(-\alpha)$ chromosome frequencies range from 0.12 to 0.23 (41–44).

Geographic variability of hemoglobin F in sickle cell disease

Fetal hemoglobin (HbF), which is composed of two α -hemoglobin subunits and two γ -hemoglobin subunits ($\alpha_2\gamma_2$), is the major hemoglobin produced during fetal development. The expression of Hb F starts early in development, peaks in mid-gestation, and by six months of age very little is expressed in most people. Fetal hemoglobin has a higher affinity for oxygen than hemoglobin A (Hb $\alpha_2\beta_2$) due to a decrease in its interaction with 2, 3-bisphosphoglycerate. It is thought that this increase in oxygen affinity allows for better oxygen transfer across the placenta from the mother to fetus.

The suggestion that Hb F may be responsible for the lack of clinical symptoms in newborns with SCD, was first made by Watson (45) in 1948, who reported that the occurrence of clinical symptoms of SCD started with a decline in Hb F levels. Since then, it is now appreciated, that Hb F is among the most important known modifiers of the clinical course of SCD (10). Fetal hemoglobin is believed to decrease hemolysis, and therefore the many complications of SCD, since both fetal hemoglobin ($\alpha_2\gamma_2$) and the mixed hybrid hemoglobin tetramers that form ($\alpha_2\gamma\beta^s$) do not participate in the polymerization of the deoxy-sickle hemoglobin. (46)

A wide range of Hb F expression has been observed among SCD patients from various parts of the world (47). Many people from the Middle East and India co-inherited additional genetic mutations that allow for the persistence of Hb F production, so that as a population they appear to have less severe disease.

There are several haplotypes of the hemoglobin S gene that correlate with region of origin of the mutation and are associated with variation in the expression of Hb F (28, 48). These include the Senegal, Saudi-Indian, Benin, and Bantu haplotypes (49). Within sub-Saharan Africa those with the Bantu haplotype have the lowest Hb F expression, while those with the Senegal haplotype the highest. Individuals with the Senegal and Saudi-Indian haplotypes have a C–T polymorphism 158 base pairs upstream of *HBG2*, one of the two γ -globin genes, that is associated with higher Hb F levels (50, 51). There is significant intra-haplotype variation and individuals homozygous for the Saudi-Indian haplotypes have higher Hb F levels than those homozygous for the Senegal haplotype (47, 52). This suggests that there are other heritable factors explaining the different Hb F levels.

Polymorphisms in the *HBS1L-MYB* intergenic region and the *BCL11A* gene correlate with Hb F levels in individuals of African descent with SCD (53–57). In the Southwestern Province of Saudi Arabia, where the large majority of SCD patients studied carried the

Benin or Bantu haplotype (96%), polymorphisms in the *BCL11A* gene are associated with Hb F levels (58). After adjusting for the *BCL11A* genotype, Saudi individuals with SCD still have significantly higher Hb F levels compared to African Americans with SCD.

These clinical and epidemiology studies complement basic science studies investigating hemoglobin gene switching. Therapeutic agents that increase Hb F expression, such as azacytidine, decitabine, butyrate derivatives, and hydroxyurea, have been investigated in clinical trials to ameliorate the complications of SCD. Hydroxyurea is the only agent yet approved for the prevention of complications of SCD (59) and polymorphisms in the *SAR1A* gene and *TOX* gene may contribute to the variability in Hb F response to hydroxyurea (60, 61).

Sickle cell disease genotype and cardiopulmonary complications of sickle cell disease

Clinical phenotypes and laboratory values vary among the Hb SS, SC, and S β^+ -thalassemia sickle cell genotypes. Patients with Hb SS have higher markers of hemolysis and lower hemoglobin values compared to those with Hb SC or S β^+ -thalassemia. Correspondingly, the prevalence of leg ulcers and priapisms, which are sickle phenotypes related to higher hemolytic rates, are more prevalent in individuals with Hb SS disease (62, 63). Other complications of SCD including stroke, vaso-occlusive pain episodes, and early mortality are also higher in Hb SS versus Hb SC or S β^+ -thalassemia (10, 64, 65). Avascular necrosis occurs at similar prevalences between the major sickle cell genotypes, although it typically presents at an earlier age in individuals with Hb SS disease (66). Conversely, the prevalence of proliferative retinopathy is highest in those with Hb SC disease followed by individuals with S β^+ -thalassemia and SS genotype (67). Comparison of the cardiopulmonary complications by sickle cell genotype suggest that individuals with Hb SS or S β^0 -thalassemia are at higher risk for acute chest syndrome, pulmonary hypertension, and low steady-state oxyhemoglobin saturations at rest compared to individuals with Hb SC or S β^+ -thalassemia (44, 68, 69).

Here we report new analyses of the PUSH and Walk-PHaSST cohorts for the relationship between three major sickle cell genotypes, Hb SS, SC and S β^+ -thalassemia, and certain pulmonary and cardiac complications of SCD. In particular, we have focused on acute chest syndrome history, oxygen desaturation, tricuspid regurgitation velocity (TRV) elevation, left ventricular (LV) size and LV function as determined by echocardiography, and N-terminal probrain natriuretic peptide (NT-proBNP) elevation. Acute chest syndrome, which includes pneumonia, is a frequent pulmonary complication of SCD (44). It is second only to vaso-occlusive crisis as a cause of hospitalization and recurrent episodes may cause debilitating chronic pulmonary disease (70). It is also a leading cause of death in SCD, accounting for approximately 25% of deaths (64, 71). The cause of acute chest syndrome is known in only about a third of cases and is related to pulmonary infections, pulmonary infarction and fat embolism (44, 72–74). Acute chest syndrome seems to be associated with a personal or family history of asthma, increased inflammatory markers, and increased phospholipase A2 levels. A physician diagnosis of asthma has been associated with increased incidence of acute chest syndrome, pain, and early death (75). In a cohort of 291 infants in the Cooperative Study of Sickle Cell Disease who were prospectively followed for 11 years, acute chest syndrome was twice as common in those diagnosed with asthma (44). Debate continues as to whether the airway hyper-reactivity reported in almost 80% of children with SCD is a distinct entity or overlaps with the approximately 20% of children diagnosed with SCD and asthma as comorbidities. A positive family history of asthma predicts increased risk of acute chest syndrome (76). Both asthma and SCD are inflammatory diseases whose severity has been associated with increases in inflammatory markers for airway and vascular

inflammation, respectively. Arachidonic acid, released from cell membranes by phospholipase A₂, produces leukotriene B₄ and cysteinyl leukotrienes. Leukotriene B₄ promotes neutrophil activation and chemotaxis. Cysteinyl leukotrienes promote bronchoconstriction, mucus production, airway edema, and smooth muscle proliferation in the lung, and also results in vascular vasoconstriction, vascular leakage and up-regulation of cellular adhesion molecules (75). The role of ventilation perfusion (VQ) mismatch in the connection between acute chest syndrome and asthma has also been a source of speculation, as has the role of nitric oxide. An increase in exhaled nitric oxide correlates with increased asthma severity, but NO bioavailability decreases with more severe hemolysis, higher plasma free hemoglobin levels, and higher TRV.

Oxygen desaturation has an independent association with left ventricular hypertrophy and diastolic dysfunction in patients with SCD (77). TRV reflects systolic pulmonary artery pressure and right ventricular systolic pressure (78). A TRV of > 3.0 m/sec in adults carries a substantial probability of finding mean pulmonary artery pressure > 25 mm Hg on right heart catheterization (79) and signifies an increased risk of early mortality (80). The clinical significance of an elevated TRV in children is less well defined, although we have reported a decline in exercise capacity over two years of follow-up in children with a TRV > 2.6 m/sec (81). Plasma concentration of brain natriuretic peptide (BNP), which is concentrated in the heart and is believed to play an emergency defense against ventricular overload in disease states (82), serves as a diagnostic marker for heart failure and left ventricular dysfunction (83, 84). N-terminal NT-proBNP is produced from proBNP cleavage (85). Serum NT-proBNP concentration correlates with pulmonary artery pressure, right ventricular dysfunction (86), and left ventricular diastolic dysfunction (87). Elevated NT-proBNP predicts a group of patients with higher mortality (86, 88, 89).

Table 3 depicts selected clinical manifestations according to three major sickle cell genotypes, Hb SS, SC and S β^+ -thalassemia, in the PUSH and Walk-PHaSST cohorts. A history of leg ulcers was higher with the Hb SS genotype than the Hb SC and S β^+ -thalassemia genotypes, but the number of severe pain episodes in the past year did not differ significantly according to these genotypes. Hemoglobin concentrations were lower and LDH concentrations, a marker of hemolysis (90), were higher in the patients with Hb SS, consistent with a greater propensity to red blood cell sickling and hemolysis. In terms of pulmonary complications, oxygen saturation <95% as determined by pulse oximetry, elevated TRV, and elevated NT-proBNP concentration were more common with the Hb SS genotype in both the PUSH and Walk-PHaSST cohorts. History of acute chest syndrome did not differ significantly according to the three genotypes.

The contributions of LV volume overload and diastolic dysfunction to mild TRV elevations seen in pediatric patients with SCD are well described (91). In the PUSH cohort, LV end diastolic Z score, an expression of LV size normalized for BSA, and LV mass index were highest in Hb SS patients; the percentage of patients with elevated LV end diastolic Z score (> 2.0) and LV mass index (> 100 g/m²) were higher than expected statistically in Hb SS patients. Systolic function as measured by ejection fraction was marginally lower in Hb S β^+ -thalassemia patients. E/Etdi, an estimate of left atrial pressure (92, 93), is used as a surrogate of diastolic function and was highest in Hb SS patients. There was a trend for a higher proportion of patients with an E/Etdi above the 95% confidence interval as defined by normal subjects (91) in Hb S β^+ -thalassemia and Hb SS patients. In the Walk-PHaSST cohort, the LV diastolic area and LV mass index were also highest in Hb SS patients while no significant differences in ejection fraction or LV lateral E/Ea were observed by sickle cell genotype.

An analysis of pulmonary function tests according to sickle cell genotype in the PUSH cohort found that patients with Hb SS or S β^0 -thalassemia had lower values for FVC, FEV1, FEF 25–75, and TLC than those with Hb SC or S β^+ -thalassemia ($P = 0.001$ in each case) and a higher value for RV/TLC (Manuel Arteta, MD; personal communication). FEV1/FVC and hemoglobin-adjusted DLCO did not differ significantly between these groups. When patterns of lung function were assigned on the basis of spirometry and plethysmography according to guidelines of the American Thoracic Society/European Respiratory Society (94), obstructive physiology was found in 19% of the patients with Hb SS or S β^0 -thalassemia and 4% of those with Hb SC or S β^+ -thalassemia. In addition, restrictive physiology was found in 9% of the patients with Hb SS or S β^0 -thalassemia and abnormal but not categorized physiology in 11%, but these abnormalities were not observed in the patients with Hb SC or S β^+ thalassemia. Thus, 39% of children and adolescents with Hb SS or S β^0 -thalassemia had abnormal pulmonary function compared to only 4% of those with Hb SC or S β^+ -thalassemia.

α -Thalassemia and cardiopulmonary complications of sickle cell disease

α -Thalassemia ($\alpha\alpha/\alpha-$ or $\alpha-/α-$) is present in approximately 1/3 of SCD patients and seems to lessen the clinical severity of disease by decreasing the mean corpuscular hemoglobin concentration, percentage of dense cells, degree of hemolysis and number of irreversibly sickled cells, and by increasing total hemoglobin levels and hemoglobin A2 levels (95, 96). There seems to be a decreased incidence of involvement of vital organs in SCD patients with alpha-thalassemia (97), and alpha-thalassemia appears to increase survival in certain populations (98). The effect of alpha-thalassemia is not always beneficial, because some complications such as aseptic necrosis and retinal disease occur more frequently in patients with concurrent alpha-thalassemia (95, 99). In individuals homozygous for the hemoglobin S mutation, α -thalassemia due to single or double α -globin gene deletions (96, 100, 101) is associated with higher hemoglobin concentrations and less hemolysis, blunted response to therapies aimed at increasing Hb F, and more frequent pain crises (27, 96, 100–106).

The effects of co-inheriting α -thalassemia with SCD on pulmonary and cardiac complications have been less clear. There have been conflicting reports on risks for acute chest syndrome in Hb SS individuals with α -thalassemia with some but not all reporting fewer episodes of acute chest syndrome in patients with α -thalassemia (96, 107, 108). Coinheritance of α -thalassemia has not been shown to be significantly associated with a TRV > 2.5 m/s in screened individuals with SCD including a subgroup analysis of Hb SS subjects (69).

Table 4 depicts new analyses of the PUSH and Walk-PHaSST Hb SS patients for selected clinical manifestations according to α -thalassemia genotype. Hemoglobin concentrations were higher and LDH concentrations were lower in the patients with α -thalassemia ($\alpha\alpha/\alpha-$ or $\alpha-/α-$) than those without ($\alpha\alpha/\alpha\alpha$), consistent with a protection from severe hemolysis and marked anemia by α -thalassemia. Among children in the PUSH cohort, number of severe pain episodes in the past year was more in the patients with α -thalassemia. In contrast, among predominantly adults in the Walk-PHaSST cohort, the number of pain episodes did not differ significantly according to the presence or absence of α -thalassemia. History of leg ulcers was lower with α -thalassemia in the Walk-PHaSST cohort. In terms of cardiopulmonary complications, α -thalassemia was associated with lower prevalence of acute chest syndrome history, oxygen desaturation, elevated TRV, and elevated NT-proBNP concentration in the Walk-PHaSST cohort, but these trends were not as clear in the PUSH cohort. In the PUSH cohort, the LV end diastolic Z score and LV mass index were higher in SCD patients without α -thalassemia while in the Walk-PHaSST cohort the LV mass index

was also higher in SCD patients without α -thalassemia. Table 5 depicts pulmonary function tests according to α -thalassemia genotype, with no significant differences being found.

Hemoglobin F and cardiopulmonary complications of sickle cell disease

Fetal hemoglobin levels vary over a 20-fold range in adults with SCD, and clinical observations suggest that higher levels of Hb F have beneficial effects (99, 109), including higher hemoglobin concentrations and less hemolysis (110–112). In one study, end organ damage seemed to be decreased in patients with Hb F levels >10% and painful crises and pulmonary complications were decreased with levels >20% (109). In the CSSCD, Hb F levels correlated inversely with the risk of painful crisis, acute chest syndrome and early mortality (10, 44, 64). A number of genetic and non-genetic factors may influence Hb F levels in patients with SCD, including age, sex, the presence of alpha-thalassemia, genetic factors linked to the beta-globin gene locus and other genetic factors (95). Despite the significant association between the level of Hb F and severity of disease, the predictive value of Hb F for early organ damage and death is less than perfect. While Platt et al (64) reported improved survival in patients with Hb F levels above the 75th percentile (>8.6%), this alone does not explain all the variability of SCD: many patients with Hb F levels above 8.6% do not have severe complications and early mortality, and many patients with Hb F levels above 8.6% have severe disease.

Table 6 depicts a new analysis of Walk-PHaSST Hb SS patients for selected clinical manifestations according to level of Hb F, divided according to levels of <10%, 10–19% and 20% or greater. The analysis was performed in patients not receiving hydroxyurea and not having undergone a recent blood transfusion, as both of these factors can substantially influence Hb F concentration. Hemoglobin concentrations were higher and LDH concentrations were lower in the patients with higher Hb F concentrations, consistent with a protection from severe hemolysis and marked anemia by Hb F. There was a trend to a lower number of acute pain episodes in the last year among patients with higher Hb F concentrations. In terms of cardiopulmonary complications, higher Hb F concentration was associated with lower prevalence of oxygen desaturation and lower LV mass index, but acute chest syndrome history, TRV elevation, and NT-proBNP concentration elevation did not differ significantly according to the Hb F categories.

Conclusion

Sickle cell disease has a high prevalence across regions of sub-Saharan Africa, the Middle East, and India and modifying factors such as SCD genotype, coinheritance of α -thalassemia, and Hb F production vary by geographic region. The impact of SCD genotype on cardiopulmonary complications appears the most consistent of these factors. Patients with Hb SS or $S\beta^0$ -thalassemia have higher rates of hemolysis reflected by higher serum LDH levels and lower hemoglobin concentrations. This has correlated with higher prevalences of leg ulcers, oxygen saturation < 95% on pulse oximetry, elevated TRV, and elevated NT-proBNP which were confirmed in the PUSH and Walk-PHaSST cohorts. There appears to be a higher prevalence of pulmonary function test abnormalities in individuals with Hb SS or $S\beta^0$ -thalassemia based on analyses from the PUSH cohort. The LV size and LV mass index were also higher in individuals with Hb SS in both the PUSH and Walk-PHaSST cohorts. It is likely that the relationship between the findings on echocardiography and clinical outcomes is complex and varies from patient to patient. Higher rates of acute chest syndrome were previously observed among Hb SS patients in the CSSCD cohort, although only a trend was observed in the Walk-PHaSST cohort. This suggests that other modifying factors may influence the pathophysiologic process for developing acute chest syndrome.

Coinheritance of α -thalassemia and increased production of Hb F correlate with lower markers of hemolysis and higher hemoglobin concentrations. The association of α -thalassemia and Hb F with cardiopulmonary complications is less clear. Coinheritance of α -thalassemia was associated with lower LV mass index in both the PUSH and Walk-PHaSST cohorts. In the Walk-PHaSST cohort, consisting primarily of adults, coinheritance of α -thalassemia was also associated with lower rates of acute chest syndrome, oxygen saturation < 95%, TRV < 3.0 m/sec, and NT-proBNP > 160 ng/L. These findings have been inconsistent in the literature and were not observed in the PUSH cohort, consisting predominantly of children. The coinheritance of α -thalassemia is associated with increased pack cell volume and the increased blood viscosity may affect blood flow and abrogate potential benefits from reduced hemolysis (113). Higher Hb F levels were associated with lower rates of oxygen saturation < 95% and lower LV mass index but no clear relationship was observed with rates of acute chest syndrome, TRV > 3.0 m/sec, or NT-proBNP > 160 ng/L in the walk-PHaSST cohort. The complex interactions between factors that influence Hb F levels in patients with SCD may influence the ability to use Hb F as a predictor for cardiopulmonary complications.

This review highlights the geographic distributions of SCD genotypes, coinheritance patterns of α -thalassemia, and varying levels of Hb F production along with influence of these factors on hemolytic parameters and pulmonary and cardiac complications of SCD. Although some of the phenotypic variation can be explained by these modifying factors, our review also emphasizes that other complex interactions including genetic modifiers need to be better understood to predict clinical severity of pulmonary disease in SCD.

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Educational Aims

1. To better understand the geographic distribution of sickle cell disease and its clinical modifiers: genotype, hemoglobin F concentration, and coinheritance of alpha-thalassemia.
2. To better understand the influence of genetic modifiers on the cardiopulmonary complications of sickle cell disease.
3. To better understand how genetic modifiers impact degree of hemolysis.

Future Research Directions

1. To identify additional modifiers that may affect risk for non-cardiopulmonary complications.
2. To identify additional modifiers that may affect risk for cardiopulmonary complications.
3. To identify specific treatments aimed at targeting modifiers of sickle cell disease to improve its complications.

Table 1

Sickle Cell Disease Genotypes in the Americas and UK

	Children and adolescents (Brazil (11)) N = 773	Children and adolescents (US- PUSH (12)) N = 504	Adolescents and adults (US, UK- walk-PHaST (13)) N = 674
Hemoglobin SS	74.4%	75.6%	74.9%
Hemoglobin SC	24.3%	17.8%	18.1%
Hemoglobin S β^+ -thal	---	3.6%	4.0%
Hemoglobin S β^0 -thal	1.3%	1.8%	1.6%
Other	---	1.2%	1.4%

Table 2

Examples of distribution of SCD genotypes in various parts of the world

	Dakar, Senegal (14) (univ. hosp.) age 1-10+ n = 556	Zaria, Nigeria (15) (univ. hosp.) age 10-52 n = 208	Ibadan, Nigeria (16) (univ. hosp.) age 3-16 n = 145	Ouagadougou, Burkina Faso (18) (urban hosp.) age 1-42 n = 387	Ouagadougou, Burkina Faso (17) (screening) age 1-30 n = 153	Greece National Registry (19) n = 1080	Riyadh, Saudi Arabia (38) (univ. hospital) age 1-69 n = 159	Andhra Pradesh, India (21) (screening) n = 78	Chennai, India (20) (urban hosp.) n = 22
Hb SS	95.7%	95.2%	87.6%	50.4%	7.8%	19.0%	71.7%	67.9%	36.4%
Hb SC	3.6%	4.8%	12.4%	49.6%	92.2%	---	---	---	---
Hb S β -thal	0.7%	---	---	---	---	81.0%	28.3% [/]	30.8%	63.6%
Hb SD	---	---	---	---	---	---	---	1.3%	---

* age given in years

[/] 26.4% Hb S β ⁰-thalassaemia; 0.2% Hb S β ⁺-thalassaemia

Table 3
Sickle cell disease genotype and pulmonary and cardiac complications in the PUSH and Walk-PHaSST cohorts

General clinical manifestations										
	>3 pain episodes PUSH	>3 pain episodes Walk-PHaSST	Leg ulcers PUSH	Leg ulcers Walk-PHaSST	Hb g/dL* PUSH	Hb g/dL* Walk-PHaSST	LDH U/L** PUSH	LDH U/L* Walk-PHaSST		
Hb SS ¹	16.8%	31.3%	1.1%	22.0%	8.5 (0.1)	8.6 (0.1)	473 (459–488)	437 (428–446)		
Hb SC ²	10.1%	27.9%	0%	9.0%	11.5 (0.1)	11.6 (0.2)	279 (260–299)	245 (235–255)		
Hb Sβ ⁺ -thal ³	17.6%	25.9%	0%	11.1%	10.7 (0.3)	11.1 (0.4)	308 (276–344)	245 (213–268)		
P	0.3	0.4	0.3	0.002	<0.001	<0.001	<0.001	<0.001		<0.001
Pulmonary complications										
	Acute chest syndrome PUSH	Acute chest syndrome Walk-PHaSST	O ₂ sat <95% PUSH	O ₂ sat <95% Walk-PHaSST	TRV 2.6 m/sec PUSH	TRV 3.0 m/sec Walk-PHaSST	BNP >160 ng/L PUSH	BNP >160 ng/L Walk-PHaSST		
Hb SS ¹	51.3%	65.3%	11.1%	21.6%	11.7%	15.0%	27.7%	25.8%		
Hb SC ²	42.7%	52.7%	0%	8.3%	1.2%	8.2%	7.0%	20.5%		
Hb Sβ ⁺ -thal ³	44.4%	64.0%	0%	0%	7.1%	0%	16.7%	8.3%		
P	0.17	0.09	<0.001	<0.001	0.013	0.007	<0.001	0.036		
Left Ventricular Size and Function										
	LV Diastolic Dimension Z score PUSH	LV Diastolic Area (cm ²) Walk-PHaSST	LV Mass Index (g/m ²) PUSH	LV Mass Index (g/m ²) Walk-PHaSST	Ejection Fraction PUSH	Ejection Fraction Walk-PHaSST	Mitral E/E _{di} PUSH	LV Lateral E/E _a Walk-PHaSST		
Hb SS ¹	1.6 (0.6–2.5)	35 (31–40)	94 (78–109)	113 (95–133)	64 (61–67)	61 (58–65)	6.5 (5.7–7.6)	6.4 (5.2–8.1)		
Hb SC ²	1.0 (–0.5– 0.7)	29 (26–33)	67 (60–78)	87 (73–108)	64 (61–66)	61 (57–66)	6.1 (5.5–6.9)	6.4 (4.9–7.6)		
Hb Sβ ⁺ -thal ³	0.4 (–0.4– 1.4)	30 (28–33)	70 (64–90)	84 (69–97)	63 (59–66)	65 (60–68)	5.9 (4.9–6.5)	5.9 (4.8–8.2)		
P	<0.001	<0.001	<0.001	<0.001	0.046	0.2	0.004	0.7		

¹ PUSH: N = 381 children; Walk-PHaSST: N = 505 predominantly adults

² PUSH: N = 90 children; Walk-PHaSST: N = 122 predominantly adults

³ PUSH: N = 18 children; Walk-PHaSST: N = 27 predominantly adults

*

In subjects without recent blood transfusion; adjusted for hydroxyurea

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In subjects without recent blood transfusion; adjusted for hydroxyurea, age and study site

In subjects without recent blood transfusion; adjusted for hydroxyurea and study site

α -Thalassemia and pulmonary and cardiac complications in hemoglobin SS subjects in the PUSH and Walk-PHaSST cohorts

Table 4

General clinical manifestations									
	>3 pain episodes PUSH	>3 pain episodes Walk-PHaSST	Leg ulcers PUSH	Leg ulcers Walk-PHaSST	Hb g/dL* PUSH	Hb g/dL* Walk-PHaSST	LDH U/L** PUSH	LDH U/L*** Walk-PHaSST	
$\alpha\alpha/\alpha\alpha^1$	14.2%	33.5%	1.3%	25.2%	8.3 (0.1)	8.5 (0.1)	498 (478–518)	459 (446–473)	
$\alpha\alpha/\alpha^-2$	23.8%	29.1%	0%	16.4%	8.9 (0.1)	9.0 (0.1)	433 (407–459)	407 (392–424)	
α^-/α^-3	22.2%	33.3%	0%	0%	9.3 (0.5)	9.1 (0.5)	392 (324–478)	302 (260–351)	
P	0.041	0.5	0.24	0.008	<0.001	0.012	0.07	0.006	
Pulmonary complications									
	Acute chest syndrome PUSH	Acute chest syndrome Walk-PHaSST	O2 sat <95% PUSH	O2 sat <95% Walk-PHaSST	TRV 2.6 m/sec PUSH	TRV 3.0 m/sec Walk-PHaSST	BNP >160 ng/L PUSH	BNP >160 ng/L Walk-PHaSST	
$\alpha\alpha/\alpha\alpha^1$	47.6%	67.0%	12.9%	25.7%	13.1%	16.6%	28.4%	28.9%	
$\alpha\alpha/\alpha^-2$	58.4%	59.6%	7.0%	13.2%	9.7%	9.6%	27.6%	19.0%	
α^-/α^-3	44.4%	40.0%	0%	0%	0%	0%	0%	20.0%	
P	0.17	0.035	0.07	0.003	0.19	0.022	0.3	0.034	
Left Ventricular Size and Function									
	LV Diastolic Dimension Z score PUSH	LV Diastolic Area (cm ²) Walk-PHaSST	LV Mass Index (g/m ²) PUSH	LV Mass Index (g/m ²) Walk-PHaSST	Ejection Fraction PUSH	Ejection Fraction Walk-PHaSST	Mitral E/E _{cd} PUSH	LV lateral E/Ea Walk-PHaSST	
$\alpha\alpha/\alpha\alpha^1$	1.5 (0.4–2.5)	34 (29–39)	90 (73–109)	109 (91–131)	64 (61–67)	62 (58–66)	6.4 (5.5–7.6)	6.4 (5.1–8.0)	
$\alpha\alpha/\alpha^-2$	0.9 (0.1–1.8)	34 (30–38)	77 (64–97)	103 (83–122)	64 (61–67)	61 (58–65)	6.3 (5.8–7.0)	6.4 (5.2–7.9)	
α^-/α^-3	0.3 (–0.4–1.2)	34 (30–34)	77 (64–80)	97 (83–129)	62 (61–66)	63 (58–66)	6.1 (5.5–6.8)	6.5 (6.0–6.9)	
P	<0.001	0.4	<0.001	0.004	0.8	0.6	0.12	0.8	

¹ PUSH: N = 234 children; Walk-PHaSST: N = 306 predominantly adults

² PUSH: N = 106 children; Walk-PHaSST: N = 147 predominantly adults

³ PUSH: N = 9 children; Walk-PHaSST: N = 10 predominantly adults

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In subjects without recent blood transfusion; adjusted for hydroxyurea

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In subjects without recent blood transfusion; adjusted for hydroxyurea, age and study site

In subjects without recent blood transfusion; adjusted for hydroxyurea and study site

Table 5

α -Thalassemia and pulmonary function tests among hemoglobin SS patients in the PUSH cohort. Results in mean (SE).

	N	FEV1/FVC	FEF25-75	TLC	RV/TLC
$\alpha\alpha/\alpha\alpha$	73	85 (0.7)	75 (2.4)	97 (2.4)	25 (1.1)
$\alpha\alpha/\alpha-$	31	84 (1.0)	74 (3.6)	98 (3.6)	25 (1.7)
$\alpha-/ \alpha-$	4	82 (2.8)	95 (10.0)	118 (9.6)	20 (4.5)
P		0.7	0.15	0.13	0.5

Table 6
Hemoglobin F and pulmonary complications in hemoglobin SS subjects in the Walk-PHaSST cohort

General clinical manifestations					
	N	>3 pain episodes	Leg ulcers	Hb g/dL	LDH U/L*
Hb F 0-9%	103	21.4%	23.3%	8.1 (0.1)	513 (493-534)
Hb F 10-19%	40	7.5%	25.0%	8.7 (0.2)	428 (395-455)
Hb F 20%+	9	11.1%	0%	10.0 (0.4)	380 (337-433)
P		0.07	0.3	<0.001	0.011
Pulmonary complications					
	N	O2 sat <95%	Acute chest syndrome	TRV 3.0 m/sec	BNP >160 ng/L
Hb F 0-9%	103	34.3%	55.3%	10.2%	16.7%
Hb F 10-19%	40	15.0%	60.0%	13.1%	23.7%
Hb F 20%+	9	0%	33.3%	11.1%	0%
P		0.003	0.6	0.7	0.8
Left Ventricular Size and Function					
	N	LV Diastolic Area (cm ²)	LV Mass Index (g/m ²)	Ejection Fraction	LV lateral E/Ea
Hb F 0-9%	396	34 (29-39)	109 (89-130)	61 (58-66)	6.4 (5.2-8.0)
Hb F 10-19%	98	34 (30-40)	105 (89-123)	62 (58-66)	6.5 (5.4-8.6)
Hb F 20%+	46	33 (28-37)	90 (81-112)	63 (60-68)	5.9 (4.7-7.0)
P		0.3	0.024	0.2	0.12

* Adjusted for site