Chapter

Inherited Disorders of Hemoglobin and *Plasmodium falciparum* Malaria

Edith Christiane Bougouma and Sodiomon Bienvenu Sirima

Abstract

An estimated 300,000 babies are born each year with severe Inherited Disorders of Hemoglobin (IDH). Despite major advances in the understanding of the molecular pathology, control, and management of the IDH thousands of infants and children with these diseases are dying due to the accessibility to appropriate medical care. In addition, as malaria has been the principal cause of early mortality in several parts of the world for much of the last 5000 years, as a result, it is the strongest force for selective pressure on the human genome. That is why, in the world, there is an overlap of malaria endemicity and IDH. Over the past twenty years several studies have shown that IDH such us hemoglobin and/or red cell membrane abnormalities confer resistance to malaria reducing hence the mortality during the first years of life. This has led to the selection of populations with IDH in malaria-endemic areas. This may explain the overlap between these two pathologies. This chapter aims to present the relationship between IDH and malaria susceptibility, make an overview of the current state of knowledge and the burden of IDH, and highlight steps that require to be taken urgently to improve the situation.

Keywords: Hemoglobin, Inherited Disorders, malaria, host genetics

1. Introduction

Despite health better care and new strategies of disease control of, mortality remains high in many countries and worldwide [1]. Malaria is the biggest cause of childhood mortality in Africa.

In 2018, malaria was the cause of about 405 000 deaths, More than 90% of these deaths occurred in sub-Saharan Africa [2].

Malaria parasites enter red blood cells during key stages of their life cycle so that there is no surprise that a change of red blood structures or make-up could affect malaria infection. Some changes of red blood cells make more resistant to malaria infection whereas others create the potential for a harmful reaction to certain antimalarial drugs [3].

Falciparum malaria has had a profound effect on human evolution, evidenced by the high frequencies of malaria protective mutations observed in populations from historically malarious regions. This big pressure has resulted in the selection of many genetic variants that confer protection against severe malaria and reducing death due to malaria in some populations [4–6].

Gene (chromosome)		Mutation		Protein	Function	Reported Genetic Associations with Malaria	Mechanistic hypotheses Proposed protective mechanism	Distribution	High Frequency
		Change	Number						
G6PD (Xq28)		Asn126Asp A376	1	G6PD	Enzyme That protects against oxidative tress	G6PD deficiency protect against severe malaria and uncomplicated malaria	Increased phagocytosis of ring-parasitised variant RBCs due to enhanced oxidative stress [94, 103]. Increased vulnerability of the G6PD deficient erythrocyte to oxidant stress causes its protection against parasitization and Reduced parasite replication in G6PD-deficient erythrocytes [94, 103].	Africa	0.4
		Val68Met G202A	1					Africa	2.5
HBA (16p13.3)	α - thalassemia (α-thal)	α + or α one gene deleted	Many	α-Globin	Component of hemoglobin	Thalassemia protects against severe malaria and severe malaria aneamia but appears to enhance mild malaria episodes in some environments	Specific protection against malaria-induced anemia [94, 103]. Reduced pathogenicity through reduced cytoadherence or resetting [86, 87]. Immunological priming through cross-species immunity between *P.vivax and P.falciparum* [74]. Increased phagocytosis of infected variant RBCs by monocytes and enhanced antibody binding and subsequent clearance of infected variant RBCs [88, 89]. Reduced resetting. Increased micro-erythrocyte count in homozygotes reduces the amount of Hb lost for given parasite density, thus protecting against SMA [43, 82, 88, 90].	Mediterranean area, South / East Asia, Pacific	0.8
		α 0 or - - both genes deleted	Many						0.03
HBB (11p15.5)	β-thalassemia (β-thal)	Many	Many	β-Globin	Component of hemoglobin	Thalassemia protects against severe malaria	Enhanced removal of parasite-infected red blood cells [96]. Reduced invasion and growth of *P. falciparum* parasites [97, 98]. Reduced pathogenicity through reduced cytoadherence or resetting [99].	sub-Saharan Africa	0.1

Gene (chromosome)	Mutation		Protein	Function	Reported Genetic Associations with Malaria	Mechanistic hypotheses Proposed protective mechanism	Distribution	High Frequency
	Change	Number						
						Enhanced antibody binding and subsequent clearance of infected variant RBCs [88]. Increased phagocytosis of ring-parasitised variant RBCs [80, 96, 100].		
S (HbS)	Glu6Val GAG/GTG	2–5	β-Globin	Component of hemoglobin	HbS alleles protect againstuncomplicated malaria and severe malaria. Reduced parasitaemia	Impairment of *P. falciparum* red cell invasion and growth under conditions of low oxygen tension [31, 43]. Enhanced removal of parasite-infected HbAS red blood cell [31, 44]. Reduced pathogenicity of *P. falciparum* infected red blood cells because of reduced expression of PfEMP1 [33, 41]. Improved acquisition of malaria-specific immunity [33, 46, 48]. Selective sickling of infected sickle trait erythrocytes leading to enhanced clearance by the spleen [49]. Reduced erythrocyte invasion, early phagocytosis, and inhibited parasite growth under oxygen stress in venous micro vessels. [49]. Enhancement of innate and acquired immunity [41]. Increased clearance of sickled infected RBCs by the spleen Acquired host immunity and increased phagocytosis of ring-parasitised variant RBCs [44]. Reduced cytoadherence and rosetting Impaired trafficking of parasite proteins to RBC surface [49]. And Inhibition of	Africa	0.2

Gene (chromosome)	Mutation		Protein Function	Reported Genetic Associations with Malaria	Mechanistic hypotheses Proposed protective mechanism	Distribution	High Frequency
	Change	Number					
					parasite growth due to oxygen-dependent polymerization of HbS [30].		
	C (HbC)	Glu6Lys GAG/AAG	2	HbC alleles protect against uncomplicated malaria and severe malaria.	Increased immune clearance of infected erythrocytes [29, 35] Impairment of *P. falciparum* red cell invasion and growth under conditions of low oxygen tension [31, 43, 47] Improved acquisition of malaria-specific immunity [33, 47, 58] Reduced pathogenicity of P. falciparum infected red blood cells because of reduced expression of PfEMP1 [33, 64, 65] and Reduced cyto-adherence of infected erythrocyte [23] increased immune clearance of infected erythrocytes [22, 29]	West Africa	0.5
	E (HbE)	Glu26Lys GAG to AAG	1–3	HbE allele reduces parasite invasion and protects against severe malaria.	Impairment of *P. falciparum* red cell invasion and growth [75]. AE heterozygotes appear to have protection from invasion into erythrocytes by *P. falciparum* malaria [53, 73]. Reduced erythrocyte invasion by merozoites, lower intra-erythrocytic parasite growth, and enhanced phagocytosis of infected erythrocytes [22, 75].	Southeast Asia	0.7

Note: The variants at the β- and α-globin loci that confer resistance to malaria and information about them, including their chromosomal location, the mutations, the protein, function and also the reported Genetic Associations with Malaria, Mechanistic hypotheses and finally their distribution.

Table 1.
Common erythrocyte variants that affect susceptibility and resistance to P. falciparum *malaria.*

The high mortality and widespread impact of malaria have resulted in this disease being the strongest evolutionary selective force in recent human history, and genes that confer resistance to malaria [7].

The history of genetics and the study of malaria are much linked. Indeed Burden of disease due to malaria across much of the world has selected for a series of traits, including the alleles of genes encoding hemoglobin, red cell enzymes, and membrane proteins.

Each year more than 7000000 babies born with either a congenital abnormality and/or a genetic disease, mainly (up to 90%) in low or middle-income countries [8].

About 25% of these births consist of five disorders, two of which, the inherited disorders of hemoglobin and glucose-6-phosphate dehydrogenase (G6PD) deficiency, are monogenic diseases [8].

In recent years there has been a major revival in scientific studies interest in the study of interactions between the inherited hemoglobin disorders and *P. falciparum* malaria, work that has been the subject of several extensive reviews [5, 9, 10].

This chapter focuses on IDH that are common enough to be of public health concern particularly those significantly associated with malaria as summarized in **Table 1**.

By presenting the relationship between IDH and malaria susceptibility, making an overview of the current state of knowledge and the burden of IDH, this chapter outline some of the more important protective genetic variants that have been identified as far as summarized in the **Table 1**. The knowledge of our understanding of the interaction between hemoglobin variants and malaria could give point to novel preventive and/or therapeutic approaches.

2. Inherited disorders of hemoglobin (IDH) and *Plasmodium falciparum (P. falciparum)* malaria

2.1 Brief review of malaria infection

Malaria is a severe infectious disease caused by parasites of the genus *Plasmodium*. *Plasmodium* is one of the longest-known parasites, which are transmitted to humans by a bite of an infected female mosquito of the species Anopheles.

Indeed, after inoculation into a human by a mosquito, the *P. falciparum* parasites enter the erythrocytic stage of their life cycle after a brief silent incubation in life (**Figure 2**). It is during this time that parasites sequentially invade and egress from their host RBCs and cause the signs and symptoms of malaria. Hemoglobin is the oxygen - carrying component and major protein of the RBC [11]. Indeed, the RBC is essential for the spread of malaria parasites, as summarized in **Figures 2** and **3**.

Despite progress towards its control of malaria, it is still the most important parasitic disease and then, one of the world's worst health problems. In 2018, about 228 million cases of malaria occurred worldwide. Most of these cases (93%) occurred in African Africa region In the same year malaria was responsible for 405 000 deaths made up to 67% (272000) of children under 5 years recognized as the most vulnerable group [12]. However, early diagnosis and fast-acting treatment prevent unwanted outcomes. Until recently it was thought that only four species of malarial parasite (Plasmodium) especially *Plasmodium. falciparum (P. falciparum), Plasmodium.vivax (P.vivax), Plasmodium. malariae (P.malariae), and Plasmoduim. ovale (P.ovale)*, have humans as their natural hosts. But, it has been found that many cases of malaria that were previously diagnosed as being due to P. malariae infection are in fact due to a fifth parasite, *Plasmodium. knowlesi (P. knowlesi)* mostly in Malaysia [13].

It has long been thought that *P. falciparum* was the only cause of severe malaria cases and deaths, until the equally destructive, if not worse, the role of *P. vivax* is

gradually highlighted and established especially in South East Asia and in Latin America [2, 14–16].

Regarding the relationship between the severity of malaria and host genetics, it appears that *P. falciparum* malaria is one of the deadly forms of malaria with a life cycle including alternatives hosts: a sexual cycle in the insect vector, an Anopheles mosquito, and a human cycle in a liver stage and an erythrocyte stage. However, the resistance mechanisms have been described in the sporozoite entry to liver cells and in the erythrocyte invasion by merozoites (**Figures 1** and **3**) [17, 18]. Genetically based resistance is involved in either altering erythrocyte invasion by merozoites, in lowering parasite growth or in impairing merozoite viability after being released from schizonts [17, 19]. The genetic resistance in the blood stage step has been extensively documented [12].

There are multiple points in the parasite lifecycle that have impacted host genetic variation, but the majority of the malaria-protective variants described so far have various important impacts on the structure and function of the RBC [2].

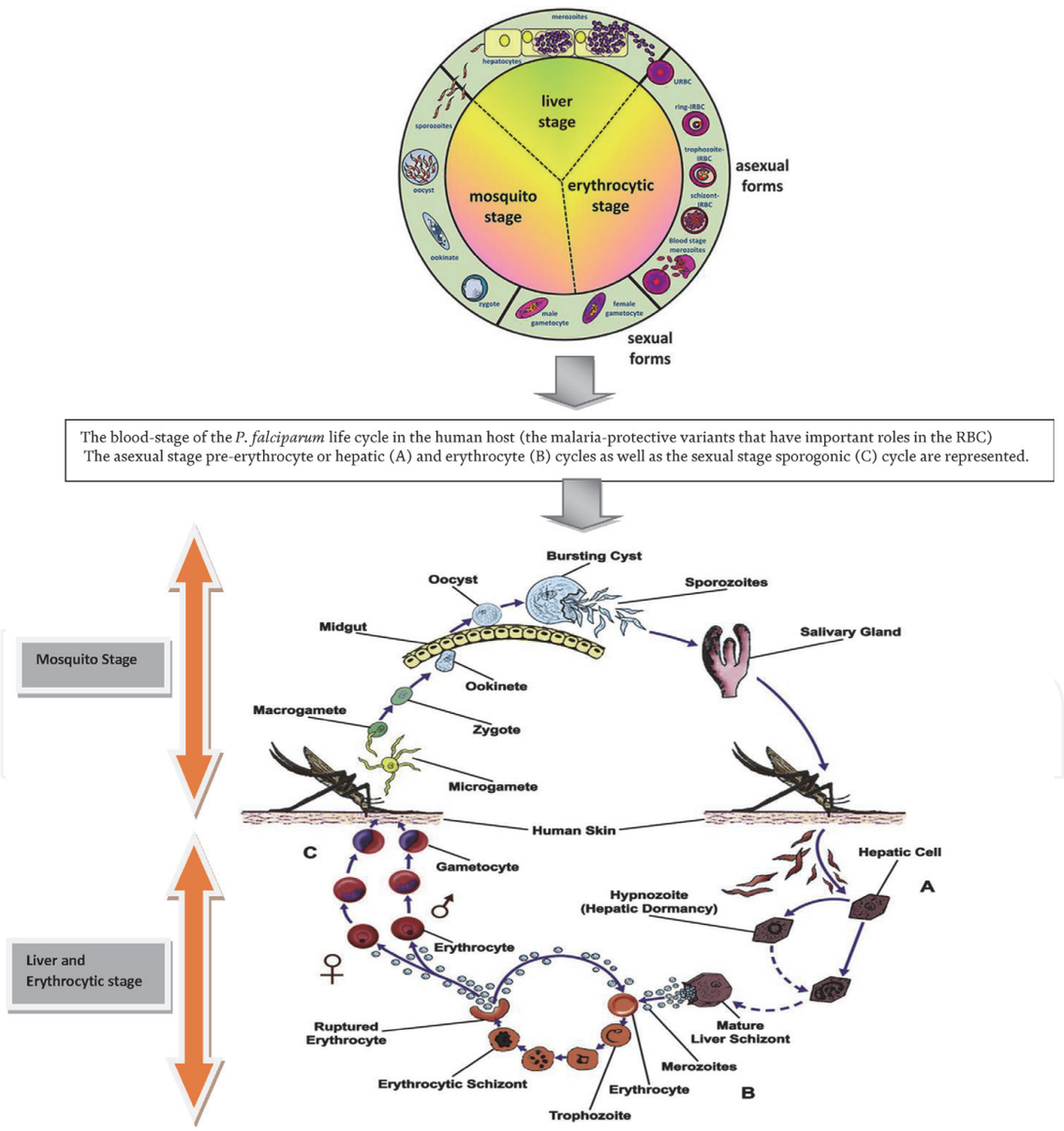

Figure 1.
The life cycle of the malaria parasite (schematic diagram illustrating life cycles of P. falciparum*, involving Anopheles mosquito and human hosts). Adapted from: Figure from Lopez et al. (2010). [Lopez C, Saravia C, Gomez A, Hoebeke J, Patarroyo MA: Mechanisms of genetically-based resistance to malaria. Gene 2010, 467:1–12.] and lee et al.(2019) [Wenn-Chyau lee, Bruce Russell, Laurent Rénia sticking for a cause: The falciparum Malaria parasites Cytoadherence paradigm immu.2019.01444].*

P. falciparum malaria is a major cause of mortality and morbidity, particularly in endemic areas of sub-Saharan Africa [2, 20]. Indeed the disease etiology is variable and is attributable to environmental factors, parasite virulence and mostly host genetics [21]. Variations in the severity of *P. falciparum* infections considered as different phenotypes include parasitaemia (hyperactive or asymptomatic), severe malaria anemia and cerebral malaria. Host genetic factors contribute to the variability of malaria phenotypes [22] and thus, should help to determine some of the mechanisms involved in susceptibility to *P. falciparum* infection. Some authors have summarized common mechanisms by which hemoglobinopathies may attenuate the pathogenesis of *P. falciparum* malaria (**Figure 2**) [11].

The knowledge gained with several studies has produced undisputed evidence about polymorphisms associated with malaria resistance. Indeed, several gene mutations and polymorphisms in the human hosts confer survival advantage and have increased in frequency through natural selection over generations. These include the classical polymorphisms that cause Sickle Cell Disorders (SCD) and haemoglobinopathies such as α-thalassaemias and G6PD deficiency and the major RCB group variants [23]. However, with news technology and experimental design, other polymorphisms have been identified that include the Dantu blood group variant, polymorphisms in the red cell membrane protein ATP2B4, and some variants related to the immune response (**Figure 3**) [10].

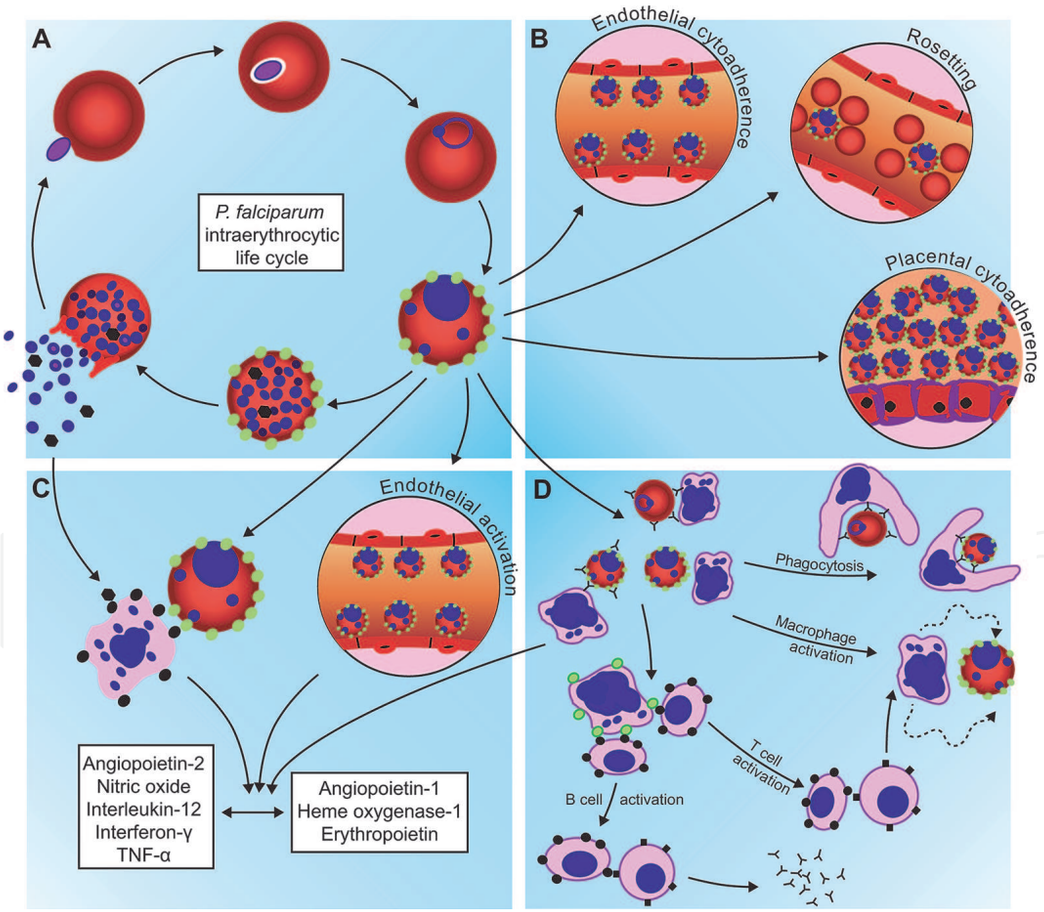

Figure 2.
Common mechanisms by which hemoglobinopathies may attenuate the pathogenesis of P. falciparum *malaria. (A) Restriction of RBC invasion or intraerythrocytic growth, thereby suppressing parasite densities in Vivo; (B) Interference with parasite-derived mediators of pathogenesis, including those involved in the binding of parasite-infected RBCs to extracellular host receptors; (C) Modulation of innate host defenses to favor protective, anti-inflammatory responses over those that drive pathogenic, pro-inflammatory responses; (D) Enhancement of adaptive cell-mediated and humoral immune responses that clear iRBCs from the blood. Source: Taylor SM, Cerami C, Fairhurst RM (2013) Hemoglobinopathies: Slicing the Gordian Knot of Plasmodium falciparum Malaria Pathogenesis. PLOS Pathogens 9(5): e1003327. https://doi.org/10.1371/journal.ppat.1003327. https://journals.plos.org/plospathogens/article?id=10.1371/journal.ppat.1003327.*

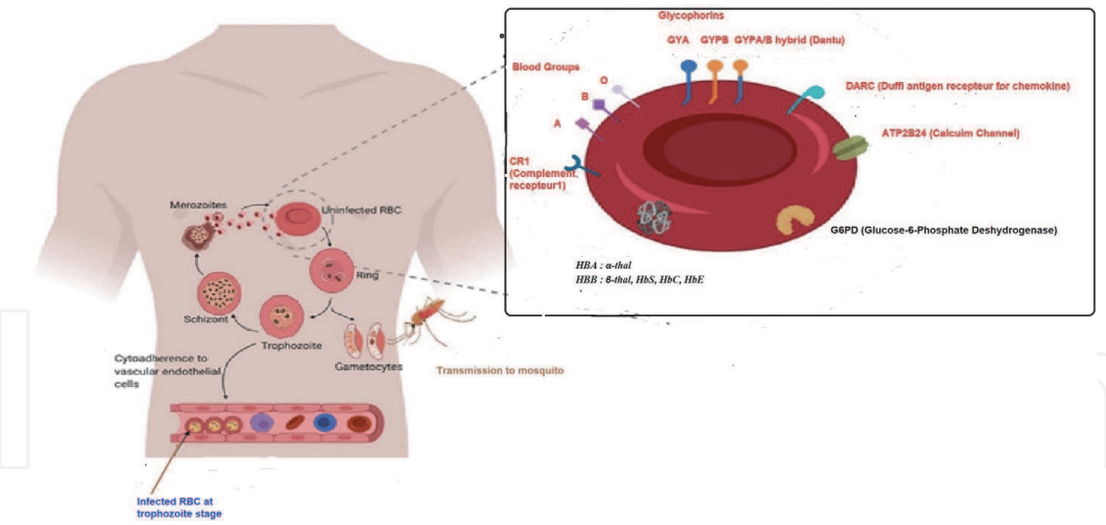

Figure 3.
Representation of the blood-stage of P. falciparum *life cycle in the human host and the malaria-protective variants that have important roles in the red blood cell (RBC). Image adapted from [1].*

2.2 Overview of inherited hemoglobin disorders

Inherited hemoglobin disorders include all disorders that are passed down through families and affect the normal properties of blood in humans. **Figure 4** summarizes the general classification IDH.

Hemoglobin disorders can be broadly classified into two general categories [24].

1. Those in which there is a quantitative defect in the production of one of the globin subunits, either total absence or marked reduction. These are called the thalassemia syndromes (quantitative disorders of globin chain synthesis/ accumulation: β-Thalassemia and α-Thalassemia

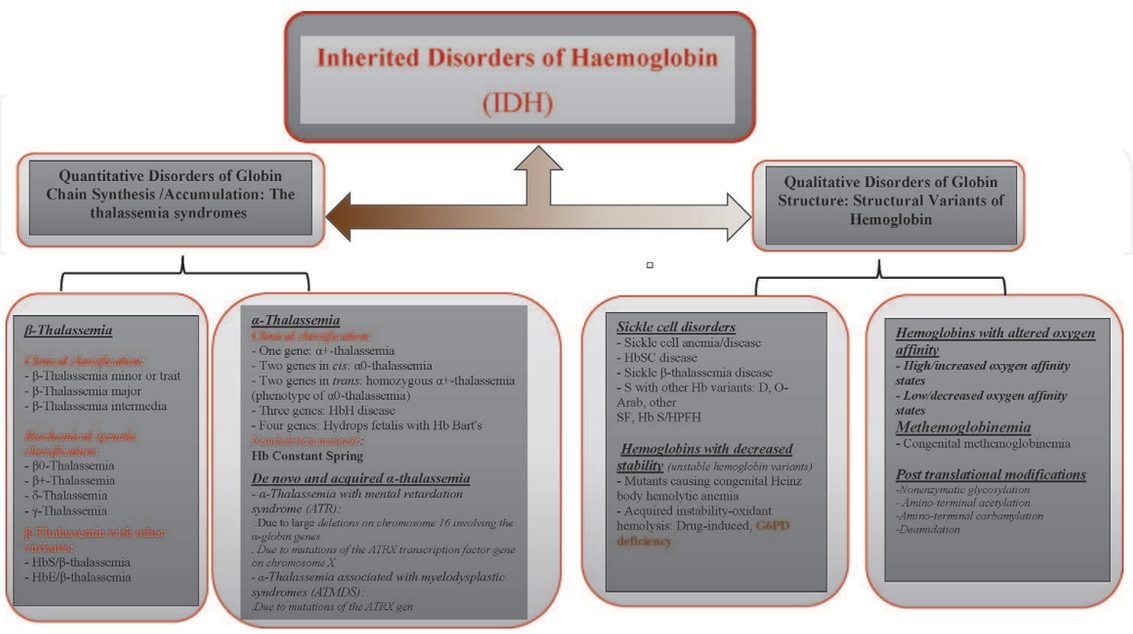

Figure 4.
Flow chart of general classification inherited disorders of Hemoglobin. Note: In this chapter we are interested in inherited hemoglobin disorders that are common enough to be of public health significance and particularly in those with a link to malaria (G6PD, α –thal, β-thal, HbS, HbC, HbE). Adapted from table of classification of hemoglobin disorders from Forget al. (2013). [Forget BG, Bunn HF: Classification of the disorders of hemoglobin. Cold Spring Harb Perspect Med 2013, 3:a011684].

2. Those in which there is a qualitative disorders of globin structure defect in one of the globin subunits: Structural variant of hemoglobin

    - Sickle cell disorders: Sickle Cell trait, Sickle cell anemia disease, SC disease, sickle β-thalassemia disease

    - Hemoglobin with decreased stability (unstable hemoglobin variants): G6PD deficiency

Hemoglobin includes four globin chains:

- fetal hemoglobin (HbF), the main hemoglobin in the fetal period which has two alpha (α) and two gamma (γ) chains (a2γ2),

- adult hemoglobin (HbA), which increases after birth up to more than 96% of total hemoglobin, has two α and two β chains (α2β2).

Human Hemoglobin genes are located in the α-globin and β-globin gene clusters in chromosomes 16 and 11. Due to spontaneous mutation, hemoglobin gene variants are present at low prevalence in all sizeable populations [5].

They fall into two broad groups structural variants that change the amino acid sequence and produce an unusual hemoglobin, [8] and thalassaemias that lower or abolish production of globin chains [12].

Morbidity and mortality rates from SCD and β-thalassemia are still very high and represent an important challenge. Increased understanding of pathophysiological aspects has lead to significant improvements in the treatment and prevention of these diseases [25]. However most hemoglobin gene variants are rare and many are harmless, but some are common because carriers are less likely than others to die from *falciparum* malaria.

We are interested in inherited hemoglobin disorders that are common enough to be of public health significance and particularly in those with a link to malaria.

2.2.1 Sickle cell disorders

SCD is a group of inherited RBC disorders; it is by far the most common IDH worldwide. SCD is caused by a variation in the gene that codes for hemoglobin, the protein in our red blood cells that helps carry oxygen to all parts of the body. The altered protein found in people with SCD is called hemoglobin S and occurs in people who have inherited the hemoglobin S (HbS), the red blood cells become hard and sticky [12].

Hemoglobin S results from an amino acid substitution at the sixth residue of the β-globin subunit: β6-Glu → Val. RBCs of persons with HbAS typically have 40% HbS and 56–58% HbA [24]. The frequency of allele S is up to 0.2 in some parts of sub-Saharan Africa [26–28]. In equatorial Africa, where malaria is endemic, the prevalence of HbAS is much higher and can reach over 30% in some populations because of the survival advantage of HbAS heterozygotes from complications of *P. falciparum* malaria. Individuals with HbAS are typically asymptomatic; severe hypoxia is required for them to experience manifestations of SCD, called sickling.

Persons who have inherited the HbS gene from only one parent are Heterozygote for the Sickle gene (AS). They carry the gene certainly, but they usually do not have the disease and are more tolerant of malaria infection, making them more likely to survive the disease [12, 29].

SCD is most common in Africa where limited resources and these resources carefully targeted are often directed towards sectors other than health. However, it

should be noted that the symptoms of SCD are often serious, substantially reducing life expectancy and often requiring intensive treatment throughout the patient's life.

Hemoglobin C results from a variation in the gene that codes for hemoglobin (β6-Glu → Lys), the protein in our RBC that helps carry oxygen around the body, It causes hemolytic anemia, splenomegaly in homozygous state and provides a degree of protection against malaria infection [12, 26]. Persons with hemoglobin C trait (Hb AC) are phenotypically normal, with no clinical evident limitations or symptoms. However, their heterozygous status, gives them a degree of protection against developing severe malaria HbC is common in malarious areas of West Africa, especially in Burkina Faso, the prevalence of HbAC is much higher and can reach over 21% [26, 28, 30, 31].

HbS and HbC caused by point mutations in the beta-globin gene, offer both substantial malaria protection. Despite the fact that the blood disorder caused by homozygosity for HbC is much less severe than that caused by homozygosity for HbS [9, 12, 26, 32], it is the sickle mutation which has come to dominate many old-world malarious regions, whilst HbC is highly restricted in its geographical distribution [33]. It is probable that this discrepancy (blood disorder between HbC and HbS) may be due to sickle cell heterozygotes enjoying a higher level of malaria protection than heterozygotes for HbC. A probable higher fitness of HbS heterozygotes relative to HbC heterozygotes could certainly have allowed the sickle cell allele to spread more rapidly. However, observations that carrying either HbC or HbS enhances an individual's capacity to transmit malaria parasites to mosquitoes could also shed light on this hypothesis [32].

Hemoglobin E results from a glutamate to lysine substitution in codon 26 (β26 Glu-Lys and GAG-AAG). Besides being a structural variant, the E variant also causes the production of an abnormal mRNA with less b-globin being synthesized. It is synthesized at a slightly reduced rate and has a homozygous phenotype similar to heterozygous β thalassemia [34].

HbE is the second commonest abnormal hemoglobin after sickle cell hemoglobin (HbS). HbE is common in South-East Asia, where its prevalence can reach 30–40% in some parts of Thailand, Cambodia and in Laos [35].

2.2.2 Thalassemia syndromes

The thalassemia syndromes are inherited disorders characterized by absence or markedly decreased accumulation of one of the globin subunits of hemoglobin. Individuals with thalassemia disease are not able to make enough hemoglobin, which causes severe anemia [24].

There is two primary types of thalassemia disease: alpha (α) thalassemias and beta (β) thalassemia disease. In the α-thalassemias, there is absent or decreased production of α-globin subunits, whereas, in the β-thalassemias, there is absent or reduced production of β-globin subunits. Thalassemias affecting the production of delta (δ)- or gamma (γ)-globin subunits are also been described but are rare and not clinically significant disorder

2.2.2.1 α-thalassemia

The α-thalassemia syndromes are usually caused by the deletion of one or more α-globin genes and are sub classified according to the number of α-globin genes that are deleted or mutated [24].

There is two primary types of α-thalassemia:

- α+ - thalassemia, in which one pair of the genes is deleted or inactivated by a point mutation,

- α 0-thalassemia, in which both pairs of genes are deleted or inactivated.

The frequency of a α thalassemia is generally 41% in regions where malaria is prevalent and in some populations, such as in Nepal, parts of India, and Papua New Guinea, it is over 80% [36]. However, in sub-Saharan African populations, a α-thalassemia frequencies do not exceed 50% despite intense malaria selection and some authors [37] suggested that this might occur because of negative epistasis with the S allele.

2.2.2.2 β-thalassemia

The β-thalassemias are characterized by a quantitative deficiency of β-globin chains, can be sub classified into those in which there is a total absence of normal b-globin subunit synthesis or accumulation. The β^{-}thalassemias are divided into two main varieties (β0-thalassemia, there is no β-chain production and β^{+}thalassemia, there is a partial deficiency of β-chain production) [24]. The molecular basis of the β-thalassemias is very heterogeneous, with over 200 different mutations having been described [38]. In general, the mutations causing β-thalassemia are point mutations affecting a single nucleotide, or a small number of nucleotides, in the b-globin gene. The frequency of carriers of β-thalassemia variants is from 5 to 20% in some areas, although not as high as the frequency of α-thalassemia variants [39].

2.2.3 Glucose-6-phosphate dehydrogenase deficiency and P. falciparum *malaria*

Glucose-6-phosphate dehydrogenase (G6PD) deficiency is a genetic disorder that results in impaired enzyme activity.

This X-linked genetic condition is characterized by reduced G6PD enzyme activity, which can remain asymptomatic. Red blood cells obtain reduced glutathione (GSH) solely from the G6PD/reduced nicotinamide adenine dinucleotide phosphate (NADH) pathway [7, 12, 30, 40, 41].

The deficiency makes red cells more susceptible to oxidative haemolysis, this disease that can cause jaundice in newborn babies and haemolytic anemia (when red blood cells break up) throughout life. This is, usually caused by an infection or exposure to certain foods or chemicals [41, 42]. One of the chemicals that can trigger severe symptoms in people with G6PD deficiency is Primaquine, the only drug currently available to clear the relapsing life stages of the *Plasmodium vivax* parasite from the liver.

G6PD and a number of other human genetic traits including sickle cell anemia and related haemoglobinopathies are predominantly found in populations living in malaria endemic countries and have been suggested to provide the host protection from severe forms of malaria [30, 43–45] and asymptomatic malaria [27].

G6PD deficiency can be common in populations with high levels of malaria infection, indeed the prevalence is even higher (8%) in malaria-endemic countries [8]. Malaria control programs need to know this to inform their policies on using Primaquine as a treatment and as a malaria control measure.

2.3 Epidemiology of inherited hemoglobin disorders and *P. falciparum* malaria

For a very long time, human beings have interacted with malaria parasites and thus the parasite has had largely time to adapt and evolve with the human host [40]. Immune processes and genetic traits have contributed to reducing the profligacy of

the malaria parasite and a wide range of genetic polymorphisms has been developed to modify the individual response to this disease. Gene mutations involved in susceptibility and resistance to *P. falciparum* malaria. It has been shown that the severity of several malaria infections (such as asymptomatic, CM and SMA) varies significantly between individuals and between populations [5]. Many of the protective variants identified thus far affect erythrocytes, where the malaria parasite spends a crucial stage of its life cycle. Several of the best studied mutations affect the globin genes encoding hemoglobin [46]. Haemoglobinopathies and G6PD deficiency are among the most common single-gene disorders, which affect RBC stability and integrity [47].

More than 700 abnormal hemoglobin have been described worldwide and more than 200 million people worldwide have RBC enzyme abnormality [48].

These genetic mutations are major causes of morbidity and mortality around the world [49]. Sickle hemoglobin and G6PD deficiency are genetically independent, their loci is located on chromosome 11 for sickle and chromosome X for G6PD deficiency genes.

Table 1 summarizes the Common Erythrocyte variant that affects malaria infection particularly *P. falciparum* malaria.

2.3.1 Sickle hemoglobin (HbS), hemoglobin C (HbC) and P. falciparum *malaria*

Sickle hemoglobin (HbS) and hemoglobin C (HbC) are both caused by point mutations in the beta globin gene, and both offer substantial malaria protection.

2.3.1.1 HbS and P. falciparum *malaria*

Heterozygosity for the sickle mutation (genotype AS) offers considerable protection against all forms of severe malaria, as well as protection against uncomplicated malaria [26, 32, 33] and parasitaemia [26, 32, 50, 51]. The different potential protective mechanisms that have been proposed and supported for sickle cell include, the growth of malarial parasites is suppressed in sickle cells [52], the abnormal display of PfEMP-1 [53], and the acceleration of acquired immunity [54]. It has also been shown that the growth rate of *P. falciparum* is retarded in HbS containing erythrocytes under conditions of low oxygen tension in vitro [55]; inhibition parasite growth by [44], miRNAs found more commonly in sickle cell trait cells than in normal cells inhibit parasite growth [56, 57].

The mechanism of the most strongly protective variant (HbS) against *P. falciparum* [1], is very complex. However studies have been shown here are two plausible mechanisms, which are not mutually exclusive, in suppression of parasite growing in red cells [55] and enhanced splenic clearance of parasitized erythrocytes [58]. Furthermore, a study summarized other possible protective effects of HbC. Indeed effects may result from [59]:

- Impairment of *P. falciparum* red cell invasion and growth under conditions of low oxygen tension [39, 55, 59]
- Enhanced removal of parasite-infected red blood cell [39, 58, 59]
- Reduced pathogenicity of *P. falciparum* infected RBC because of reduced expression of PfEMP1 [53, 60, 61]
- Improved acquisition of malaria-specific immunity [60, 62–64]
- Inhibition of parasite growth due to oxygen-dependent polymerization [37]

- Selective sickling of infected sickle trait erythrocytes leading to enhanced clearance by the spleen. Reduced erythrocyte invasion, early phagocytosis, and inhibited parasite growth under oxygen stress in venous micro vessels [65].

- Enhancement of innate and acquired immunity [53]

- oxygen-dependent polymerization of HbS is responsible for *P. falciparum* growth inhibition [66]

A study with children (between the ages of 2 and 10 years) found that the protective effect of HbAS against malaria increased from 20% to 56%, which implies that it enhances or acts in synergy with the acquired immune response [54, 67].

The compromise between risks and benefits allows us to maintain the HbS polymorphism at allele frequencies of environ 10% in many parts of Africa, despite the lethal consequences for homozygotes, which provides the most striking known example of heterozygote advantage in human genetics [9].

- Severe *P. falciparum* malaria

Some case–control and prospective cohort studies [10, 33, 54, 68–71] indicate that HbAS is consistently associated with large reductions in the risk of severe malaria.

- Uncomplicated *P. falciparum* malaria

A comparative studies [33, 44, 71–73] and several prospective studies [5, 26, 37, 74] have shown the reduction of risk in malaria attributable to HbS. In fact, the HbAS genotype protects against uncomplicated *P. falciparum* malaria by about 30% [33, 71]. In addition, this has been further confirmed by some genome-wide association studies [70, 71].

- *P. falciparum* parasitaemia

Cross-sectional studies have reported conflicting data on the prevalence of *P. falciparum* parasitemia in asymptomatic HbAS children compared with HbAA children.

A lower prevalence of parasitaemia in HbAS children was reported in some studies, [6, 26, 75], when others studies found contrary results of similar prevalence [72, 76, 77] or of higher prevalence [78, 79].

In these surveys, parasite densities were reported in HbAS children as lower [6, 62, 76, 79, 80] or similar [27, 78, 81, 82] to those in HbAA children. We can conclude that HbAS does not consistently protect from *P. falciparum* parasitaemia.

2.3.1.2 HbC and P. falciparum *malaria*

A recent meta-analysis concluded that homozygotes for βC (Hb CC) were strongly protected against severe malaria, and heterozygotes (HbAC) were mildly protected [10, 33]. It has also been found that both both heterozygotes and homozygotes of HbC are protected against severe malaria [26, 30, 31, 44, 83] but the protective effect appears to be substantially greater in homozygotes [44].

Although a cohort study in Mali reports an increase in the incidence of clinical malaria in AC individuals relative to AA [84].

HbC genotypes are not fully elucidated [56], but several mechanisms have been proposed to explain the malaria protection offered by HbC [32], including abnormal

intra-erythrocytic development of the parasite leading to lower *P. falciparum* replication rates in subsets of CC erythrocytes [65]; *abnormal P. falciparum* erythrocyte membrane protein 1 (PfEMP-1) display, leading to reduced cytoadherence and possibly reduced parasite sequestration [85], and accelerated acquisition of immunity against malaria [63]. In addition, the protective effect of HbC may result from:

- Increased immune clearance of infected erythrocytes [36, 46].
- Impairment of *P. falciparum* red cell invasion and growth under conditions of low oxygen tension [39, 55, 63]
- Improved acquisition of malaria-specific immunity [60, 63, 78]
- Reduced pathogenicity of *P. falciparum* infected red blood cells because of reduced expression of PfEMP1 [60, 85, 86] and Reduced cyto-adherence of infected erythrocyte [30]
- increased immune clearance of infected erythrocytes [28, 36]
- Besides, on observations of reduced parasite cytoadherence abnormal PfEMP1 expression, clustering of erythrocyte band3 protein, and altered surface topography of the erythrocyte membrane in the presence of HbC, it would appear that the protective effect of HbC works by increasing the immune clearance of infected erythrocytes [85, 87, 88].

2.3.1.3 Severe P. falciparum *malaria*

Compared to healthy children, HbC appears to protect against severe malaria to a lesser degree than HbS and in proportion to allele frequency [31, 33, 44, 73, 89]. Protection from specific severe malaria syndromes has not been fully investigated in HbCC; in one study [90] HbAC showed mild protection from cerebral malaria (CM) and severe malarial anemia (SMA). When compared to children with uncomplicated malaria, protection from severe malaria is inconsistent: non-significant protection is reported from severe malaria in some studies [30, 33, 44, 69] of HbCC and HbAC, and from SMA in other studies [30, 33, 69] that combined homozygotes and heterozygotes. Significant protection from CM was reported in one study of Malian children that combined homo- and heterozygotes [30, 33] Prospective studies have not reported the incidence of severe syndromes in HbC children. Thus, convincing evidence for protection from severe malaria owing to HbC derives largely from few case–control studies. Also, a further strong evidence for overall protection comes from a recent GWAS, which concluded that for each copy of the HbC allele, the risk for severe *P. falciparum* malaria was reduced by 29% [56, 70].

- Uncomplicated *P. falciparum* malaria

Few studies have reported the risk of uncomplicated malaria associated with HbC. However some comparative studies [44, 72] and prospective studies have yielded conflicting results [26, 33, 74, 91]. Further studies are still needed to show the evidence of protection from uncomplicated malaria afforded by HbCC and HbAC.

- *P. falciparum* parasitaemia

In most studies cross-sectional surveys with adults and children, HbC has not been associated with a reduced prevalence of *P. falciparum* parasitaemia

[76, 78, 80, 92, 93] or *P. falciparum* density [76, 78, 80, 94]. The incidence of asymptomatic parasitaemia did not differ between HbAC and HbAA children in Mali study [26, 74]. However, in Burkina Study HbAC genotype was associated with a lower incidence of clinical malaria relative to AA among children. Thus, HbC does not appear to modify the risk of *P. falciparum* parasitaemia.

2.3.1.4 HbE and P. falciparum *malaria*

HbE is an extremely most common structural hemoglobin variant that occurs at high frequencies throughout Southeast Asia and has reached an allele frequency of up to 70% in some areas of northern Thailand and Cambodia [52]. It is a β-hemoglobin variant, which is produced at a slightly reduced rate and hence has the phenotype of a mild form of β thalassemia [95].

Generally, none of HbS or C variants are present in Southeast Asia and HbE is in general also absent from populations in which HbS and HbC are present [52].

HbE is an extremely most common structural hemoglobin variant that occurs at high frequencies throughout Southeast Asia and has reached an allele frequency of up to 70% in some areas of northern Thailand and Cambodia [34].

AE heterozygotes appear to have protection from invasion into erythrocytes by *P. falciparum* malaria [4, 65, 72, 96]. Moreover, the protective effect of HbE may result from impairment of *P. falciparum* red cell invasion and growth [96], lower intra-erythrocytic parasite growth, and enhanced phagocytosis of infected erythrocytes [28, 96].

When the frequency of HbE is high, some other red cell disorders, such as a-thalassemia, can be also in high frequency. Although extensive sequence analysis has not been carried out. [97]. However, the E allele found in China is on the same haplotype as that found in Thailand [98], suggesting that it does not have a different origin.

Few studies have been done to characterize the mechanisms of malaria protection. Three categories of effects are relevant: reduced parasite growth and development, altered adhesion of parasitized RBCs to endothelium, and impact on the immune system. In vitro studies of HbEE and HbAE RBCs have found reduced invasion and growth of HbE [96, 99]. Clearly, more work needs to be done to answer further questions about the protective impact of HbE.

- Severe *P. falciparum* malaria

Meta-analysis of few studies [21, 33, 100] that compared the prevalence of HbE in severe and uncomplicated malaria cases demonstrated no evidence of protection, though this should be interpreted cautiously given the significant.

Considering heterogeneity of the findings and the highly selected settings of the studies, more investigations are necessary to conclude on possible protection of HbE.

- Uncomplicated *P. falciparum* malaria

We have not identified studies that have quantified clearly susceptibility to malaria by HbE.

- *P. falciparum* parasitaemia

A cross-sectional study conducted in India reported a significantly lower prevalence of *P. falciparum* parasitaemia in patients with HbE (AE or EE) compared with patients with HbAA [101].

2.3.1.5 Hemoglobins S/C and malaria transmission

Some studies suggest that human genetic variation at the β-globin locus can influence the transmission of malaria. Indeed the same genetic variants that are protective against infection also showed an association with the intensity of malaria transmission. Hemoglobin variants C and S protect against severe malaria but their influence probably on parameters not directly linked to disease severity such as gametocyte carriage and infection chronicity. Moreover, some studies provided evidence that hemoglobin variants selected for the protection against malaria might also have a broader impact on local epidemiology by influencing the frequency of parasite, including the carriage of gametocytes [3, 32].

2.3.2 Thalassemia (α and β) and P. falciparum malaria

The thalassemias are the most common Mendelian diseases of humans and constitute a major global health problem [39]. This is a group of clinical disorders that result from defective production of α- or β-globin chains, which arise from deletions or other disruptions of the globin gene clusters on chromosomes 11 and 16 [9].

A study in Kenyan children found that both heterozygous and homozygous a1 thalassemia was protective against severe malaria [102], whereas a study in Ghanaian children found that heterozygotes were protected [103]. However, a study conducted in Papua New Guinea, founded the risk of severe malaria (other childhood infections) was reduced by 60% in children who were homozygous for α1 thalassemia and to a lesser degree in heterozygotes [104]. The protective mechanism of thalassemia is not well known. Flow-cytometry studies in vitro have shown that erythrocytes with the thalassemia phenotype show reduced parasite growth [105] and increased binding of antibodies from malaria-immune [106].

2.3.2.1 α - thalassemia

The distribution of both a and b thalassemia variants seems to correspond closely to the regions that have historically had high rates of malaria [23] and the local distribution of these variants also corresponds to endemic malaria [36, 107]. Several studies have shown protection from severe malaria for individuals with a α-thalassemia, compared with individuals without thalassemia [37, 103, 104]. In addition, some authors. In a case–control study have shown protection from severe malaria for a α heterozygotes and homozygotes compared to normal aa/aa genotype [7, 37]. Overall, it appears that.

many haplotypes that reduce the expression of α-globin provide a selective advantage in resistance to severe malaria. Indeed some mechanisms have been proposed to explain the malaria protection offered by α thalassemia:

- Specific protection against malaria-induced anemia [90, 104]
- Reduced pathogenicity through reduced cytoadherence or resetting [108, 109]
- Immunological priming through cross-species immunity between *P. vivax and P. falciparum* [94]
- increased phagocytosis of infected variant RBCs by monocytes and Enhanced antibody binding and subsequent clearance of infected variant RBCs [110, 111]

- Increased micro-erythrocyte count in homozygotes reduces the amount of Hb lost for given parasite density, thus protecting against SMA [55, 104, 110, 112]

- Severe *P. falciparum* malaria

Some studies [90, 102–104] investigated α-thalassaemia showed protection against severe malaria, malarial anemia and additionally, protection from cerebral malaria [94].

- Uncomplicated *P. falciparum* malaria

Several prospective studies have assessed the incidence of uncomplicated malaria in α-thalassaemic children, with conflicting results. Indeed some studies showed the incidence of falciparum malaria was higher in α-thalassemia homozygotes and heterozygotes [94]; in contrast, other studies, found a lower incidence [37, 113]. However, other studies have found no protection for both homozygotes and heterozygotes

- *P. falciparum* parasitaemia

In cross-sectional studies, α-thalassaemia was not associated with the prevalence of parasitaemia [79, 103, 112, 114, 115]. In prospective study of children conducted in Papua New Guinea, both α- thalassaemia homozygotes and heterozygotes had fewer episodes of PCR-detectable parasitaemia than those without α thalassaemia, [115, 116] though this outcome has not been investigated in other studies.

Finally, there is no evident data to confirm a protective effect of α-thalassaemia against asymptomatic parasitaemia [112, 114].

2.3.2.2 β–thalassemia

Haldane has explain the very high level of β-thalassemia in some Mediterranean populations by the 'malaria hypothesis' of Haldane [117].

There is ordinarily only one copy of the HBB gene and βþ and β0 thalassemia showing the reduction and loss, respectively, of the production of functional protein. Individuals with α-thalassemia major, have profound anemia while Heterozygotes typically have mild anemia, however, symptoms can vary greatly in severity from having severe anemia to being a symptomless carrier. [118]

Generally, β-thalassemia is more of a public health problem because of this higher morbidity than α-thalassemia.

Several mechanisms have been also proposed to explain specific protection against malaria-induced

- Enhanced removal of parasite-infected RBC [45]

- Reduced invasion and growth of *P. falciparum* parasites [119, 120]

- Reduced pathogenicity through reduced cytoadherence or resetting [121]

- Enhanced antibody binding and subsequent clearance of infected variant RBCs [110]

- Increased phagocytosis of ring-parasitised variant RBCs [45, 102, 122]

- Severe *P. falciparum* malaria

To our knowledge, no studies have investigated the risk of severe malaria in patients with β-thalassaemia.

- Uncomplicated *P. falciparum* malaria

A case–control study in Liberia, have showed a low prevalence of β-thalassaemia in participants with uncomplicated malaria compare to community controls [72].

- *P. falciparum* parasitaemia

A other study that was done in Northern Liberia, found no differences, although they did report lower parasite densities in those with β-thalassaemia [46].

2.3.3 *Deficiency Glucose-6-phosphate dehydrogenase (G6PD) and* P. falciparum *malaria*

G6PD deficiency is a common X-linked recessive genetic disorder inherited from parents. Although, in most cases, G6PD-deficient individuals appear normal, it can lead to life-threatening anemia in severely G6PD-deficient individuals during oxidative stress-induced by the consumption of certain foodstuff (fava beans), legumes, and taking such as particular antimalarial (primaquine and pamaquine), sulfonamide, sulfamethoxazole, and other drugs and chemicals [123] and also probably infection with microorganisms [124]. Additionally, some authors show that G6PD deficiency increases the risk of severe neonatal hyperbilirubinemia, which can lead to lifetime disability with kernicterus if inadequately treated [115, 125].

However, there is a big beneficial effect of G6PD deficiency. Some studies have reported that G6PD deficiency provides resistance against malaria as the malaria parasite cannot complete its life cycle in compromised G6PD deficient RBC which have a decrease in life span or because of early phagocytosis of deficient RBC [126, 127]. Deficient G6PD enzyme activity has been shown to correlate with protection against severe malaria [43, 89]. Reduced parasite replication in G6PD-deficient erythrocytes is thought to be the mechanism of protection [128], but the parasite appears to counter this by manufacturing G6PD itself [129].

The geographical distribution of G6PD deficiency is consistent with evolutionary selection by malaria [130], and a hypothesis of positive selection [131–133]. The results of studies examining the risk of malaria for various G6PD-deficient genotypes are not consistent. Some authors [43, 134] found in Gambia and Kenya that the reduction in risk of severe malaria in male hemizygotes was 58% and that the reduction in risk for heterozygous females was 46%. Other hand, other authors found that in two populations in Mali, the reduction in risk of severe malaria in male hemizygotes was also 58%, but no reduction in risk in the female heterozygotes [93]. Some authors [27, 133] found no protective effect for either male hemizygotes or female heterozygotes. However, a protective effect has been reported on for females that were found to be G6PD deficient. This finding appears to be based on the incomplete correlation of genotype and phenotype for G6PD deficiency in female heterozygotes due to variable inactivation of the two X chromosomes [127].

Several mechanisms have been proposed to explain the malaria protection offered by G6PD deficiency [45, 127].

- Increased phagocytosis of ring-parasitized variant RBCs due to enhanced oxidative

- Increased vulnerability of the G6PD deficient erythrocyte to oxidant stress causes its protection against parasitization

- Reduced parasite replication in G6PD-deficient erythrocytes

2.4 Global burden of the hemoglobin disorders

Hemoglobinopathies are a group of IDH initially described in the subtropical regions, they are now spread all around the world. Their high frequency and clinical severity make them a global health burden mostly in Africa where there is a huge lack of resources. The measure of the yardstick of under-5 mortality has been used to assess the broad effect of hemoglobin disorders on health because most affected children can die in early childhood and most survivors can have a chronic disease. Some authors show that the disease may be cause of at least 3.4% of deaths in children aged under 5 years [135]. However, it is very difficult to estimate the burden especially as inherited disorders affect families and then communities. Worldwide, over 1% of couples are at risk for IDH most have at least one affected child. Most affected children could die in early childhood although there are now better health facilities and medical care.

Although the West African death rate in children aged under 5 years is 18.4%, This rate is 16.5% for children born to couples, not at risk for sickle-cell disorders, and 40% for children born to couples who are at risk [135]. Clearly, methods to assess the health burden of inherited disorders must include also a family perspective [135].

The burden of disease due to malaria across worldwide vary according to selected visible traits of major medical importance, including the alleles of genes encoding hemoglobin. There are several reasons for the extremely high frequency and uneven distribution of inherited hemoglobin disorders. Natural selection is by far the most important, because of the frequency of the heterozygote and the protection against malaria afforded to the homozygotes of thalassemia and HbC, followed by consanguineous marriages [9, 136]. The epidemiological transition whereby, owing to improvements of health care services, nutrition, and health positive social and behavioral factors, babies who would have probably died from the more severe hemoglobin disorders survive nowadays [39]. Then, the migration from areas of high frequency of SCD into regions like Europe and the United States are also cited.

Currently, there are only limited data on the gene frequencies and the number of births of patients with common hemoglobin disorders, particularly in Africa. Micro mapping studies involving many different centers in these countries have recently found that there is remarkable diversity in the frequency of the hemoglobin disorders even over small geographical distances [137–139].

For the future more micro mapping data are then needed to provide an accurate picture global burden according selective factors distribution [60]. Hemoglobinopathies are so common that they provide a convenient model for working out a genetic approach to the control of chronic childhood diseases. At present, about 250 million people (4.5% of the world population) carry a potentially pathological haemoglobinopathy gene. Haemoglobinopathy control programs, based on WHO approaches and recommendations, have been established in different countries in all WHO Regions and have been successful in the management of the problem by reducing the burden of the Hemoglobin Disorders [108].

Nowadays effective prevention programs have been carried out successfully in many developed countries concerning medical care for hemoglobinopathies. The programs should be extended and followed to African regions where hemoglobin

disorders are frequency very high. Indeed this frequency accounts for more than 70% of total hemoglobinopathies in the world [140]. Reducing the incidence of IDH, better prevention against IDH should remain the major priority of health services in order to reduce the burden of heamoglobinopathies.

2.5 Global burden of the Malaria

Despite the global awareness with the promulgation of malaria eradication goals, despite the colossal efforts deployed in many forms: international and bilateral cooperation, foundations and humanitarian agencies, philanthropic works. Malaria remains a crucial public health concern within the world in general and in tropical countries in particular. According to the latest Global Malaria Report, there were 228 million cases of malaria in 2018, of which there were 405,000 deaths. Almost all of the morbidity and associated lethality, respectively 93% and 94%, occur in Africa (WMR, 2019) more than 50% of the disease burden of malaria is borne by only 6 countries namely: Nigeria (25%), the Democratic Republic of Congo (12%), Uganda (5%) and Côte d'Ivoire, Mozambique and Niger (4% each). With 67% of malaria-related deaths, children under 5 remain the most vulnerable group [2]. These health consequences of malaria bring with them very important economic and social tragedies. In some cases, the resulting disruption of family structures has consequences for the family itself and for the whole community over several generations [141]. The incidence of malaria is inversely proportional to the level of development of the affected societies. The global distribution of gross domestic product per capita shows a strong correlation between malaria and poverty. Malaria generates direct costs (medical care) and indirect costs (loss of productivity linked to disabilities and deaths due to malaria) [141]. The relief of all this sharp and stifled pain requires strengthening malaria prevention programs and promoting multidisciplinary research on effective and safe antimalarial drugs and vaccines.

3. Discussion

This review confirms that the malaria parasite has co-evolved with its human host, each struggling for survival. The resulting stigmas appear as polymorphisms of the human genome. This process resulted in a symbiotic association, conferring to the host a relative protection against parasitic infection on one hand and allowing the parasite a greater longevity and proliferation due to host acquired resistance on other hand About polymorphisms of the proteins of red blood cells conferring an innate relative resistance to malaria, it is established that with an effect size >80%, the HbS variant confers the strongest protective effect against severe malaria, while the α- thalassemia confers a protective effect of about 40% in homozygotes [70, 71].

Other genes such as G6PD deficiency have also been shown to be present at high frequencies in endemic malaria populations [9, 10, 22, 142]. However, taking into account the limited mapping specific gene variants of early studies, recent studies have shown that the significant effects of known candidate genes would explain only a small fraction of the heritability of malaria [10, 22, 63, 143, 144]. This indicates that the genetic architecture of susceptibility to malaria is much more complex and that "missing heritability" could be explained by polygenetic or epigenetic effects or by gene–gene and gene–environment interactions [10, 145].

The MalariaGEN consortium has greatly contributed to understanding the correlation between the pathogenesis of malaria and polymorphisms in human RBC.

A better understanding of how changes in RBC physiology affects malaria pathogenesis may uncover new strategies to combat the disease. Understanding the

molecular basis of these polymorphisms may shed additional light on the variation in human susceptibility to malaria and offer insight into mechanisms of malaria pathogenesis likewise, a better understanding of red cell membrane structure and function will offer opportunities for the discovery of new and urgently needed therapeutic targets for the treatment of malaria.

4. Conclusions

Hemoglobinopathies are among the most common monogenic diseases in populations. The complexity of their pathophysiological processes, the severity and diversity of their clinical manifestations reflect the relevance of their scientific interests. Genetic polymorphisms that affect the structure and production of the β- or α-chains of hemoglobin are variously associated with protection from a range of clinical manifestations of *P. falciparum* infection. The degree of protection conferred by hemoglobinopathies, in general, is greatest against severe malaria, moderate against uncomplicated malaria, and probably absent against asymptomatic *P. falciparum* parasitaemia. Therefore, there is a positive relationship between the frequency of either βC or βS and malaria selection intensity favors.

However, people with hemoglobin disorders could be high-risk groups. Indeed subjects admitted with malaria are twice more likely to die than those admitted for other pathologies. The screening and genetic counseling for hemoglobin disorders should be an intrinsic part of health care in most countries. Health facility's services should be designed to provide a foundation for more comprehensive community genetics services because hemoglobin disorders are commonly a point of entry for genetic approaches into health systems.

Although information about the precise world distribution and frequency of the inherited hemoglobin disorders is still limited, there is no doubt that they are going to pose an increasing burden on global health resources in the future. Increased knowledge of the biological basis of these diseases would offer significant advances in their therapeutic management and in the prevention of the occurrence of new cases. Indeed the high frequency of IDH because of natural selection associated due to consanguineous marriages in some countries could be reduced through public awareness campaigns. Improving health care conditions in general and those related to pathologies associated with IDH would enhance affected children's survival. Hemoglobin variants could shape the distribution of malaria parasites in human populations and their transmission potential. Therefore, the knowledge of our understanding of the interaction between hemoglobin variants and malaria parasites is still being incomplete even if it has improved these last years. Nonetheless, with the interest in malaria elimination, knowledge on how these prevalent genetic variants influence parasite distribution and probably cumulative host transmission potential would be particularly valuable and necessary.

The malaria parasite has co-evolved with its human host in a struggle for their survival. The scars of this war on the human genome are polymorphisms conferring an innate resistance to malaria. Regarding relationships between malaria and human genetic alterations of RBC proteins, it appears that the MalariaGEN studies have clearly opened new doors to understand the malaria burden on human RBC polymorphisms and thus malaria pathogenesis. These new pieces of knowledge will help to redefine or readjust malaria control strategies.

In fact, despite the complexity of these interactions, hemoglobin variants in general and hemoglobinopathies, in particular, show a good model and natural experiment identifying cellular and molecular mechanisms by which *P. falciparum* produces morbidity.

Vaccines are unquestionably the most cost-effective way for malaria control. The new generation of vaccine delivery systems is increasingly moving towards co-administration of certain immunostimulants and the use of more than one antigen in the same system. In any event, the best vaccine should be effective, safe, low cost, available, and easy to administer.

5. Perspectives

WHO has announced an ambitious goal of global malaria elimination by 2030. Malaria elimination is possible but will require adaptive and well-managed programs and the implementation of evidence-based surveillance strategies and strong national responses, with adequate funding and human resources.

Over the past 20 years, significant progress has been made in the fight against malaria worldwide, with impressive reductions in transmission in many endemic regions. However, the successful outcome of the global malaria eradication research program will depend on the development of new and more effective tools, including rapid diagnostic tests, drugs, vaccines, insecticides, and awareness raising. Nowadays, genetic and genomic knowledge of malaria parasites, vectors (mosquitoes), and human hosts are available. These bits of knowledge could and should be used for the development of the latest generation tools that are more efficient and secure. The situation is all the more urgent and worrying as there is the emergence of a resistance of *Plasmodium falciparum* to the first-line drug, namely artemisinin.

In any event, the development of an effective, safe and operational malaria vaccine remains the ultimate objective that we must achieve. In this perspective, we must watch and take into account the genetic variation of the parasite population, which threatens to undermine our efforts.

A big significant milestone in scientific advancements of the last twenty years was already the elucidation of the genomes, transcriptomes, and proteomes of many pathogens, including malaria parasites. These informations give a clearer and more detailed picture that provides the foundation for new approaches to refine existing targets as well as to identify new target antigens for the development of more efficient and effective vaccines, drugs and diagnostic tests. It is clear that further progress is needed for the development of a malaria vaccine, based on basic research, in order to identify new target antigens and better understand how different adjuvants will affect balance, sustainability, and the effectiveness of the responses. To hope for an effective vaccine, emphasis should be placed on mixtures of antigens combined with potent adjuvants, not only to induce the necessary effective responses, but also to increase the possibility of inducing at least partial cross-immunity by including a range of Plasmodium epitopes.

Innovative genome-based vaccination strategies have shown the potential of a number of pathogens, including malaria. A rational genome-based vaccine design, allowing the selection of the best possible targets by prioritizing antigens according to clinically relevant criteria (frequency and magnitude of the clinically relevant immune response and/or biological function), will overcome the problem of poor immunogenicity and poor vaccine protection that have undermined the development of malaria vaccines in the past 30 years.

The use of gene drive technology could revolutionize ecosystem management. These emerging technologies with potential global effects are being offered to researchers and now subject to public discussions regarding environmental and safety concerns. The relative protection against malaria that heamoglobinopathies would confer, justify justly that we are still investigating this correlation to determine all its nature and its power. Despite this relative advantage over the

manifestations of malaria in subjects with sickle cell trait, gene therapy is a new approach to healing patients with hemoglobinopathies, which must be popularized. In this sense, clinical trials are underway with promising results. However, there are still frontiers to explore that could improve this approach: the stoichiometry between transgenic hemoglobin and endogenous hemoglobin in relation to the different genetic mutations of globins; the supply of donor cells, such as the use of induced pluripotent stem cells (iPSC); and the use of safer gene insertion methods to prevent oncogenesis.

The overall prospects for malaria elimination are clear, encouraging and the potential opportunities are endless. Technological discoveries and advances are happening at an incredibly fast pace. The context of discovery and use of new technologies makes the eradication of malaria within the reach of the test tube. However, regardless of the time it will take, we must increase our efforts; intensify networking, with the financial support and strong political will of our leaders, especially in Africa where the problem of malaria is the most important.

Acknowledgements

We would like to acknowledge all authors whose articles were cited in this chapter.

Conflicts of interest

The authors have declared no conflict of interest.

Authorship contributions

BEC wrote the manuscript. SBS read and approved the final manuscript and agree to be accountable for all aspects of the work.

Contributor information

Bougouma Edith Christiane, Email: eddy.cnrfp@gmail.com/e.bougouma@gras.bf. Sirima Bienvenu Sodiomon, Email: s.sirima@gras.bf/gras@fasonet.bf

Funding statement

The authors received no specific funding for this work.

Competing interests

The authors declare no competing financial interests.

Ethics approval

There is no ethical approval for this article.

Disclosures

None.

Acronyms and Abbreviations

Acronyms

INSP	Institut National de Sante Publique
GRAS	Groupe de Recherche Action en Santé

Abbreviations

INSP	Institut National Sante Publique
GRAS	Groupe de Recherche Action en Santé
IDH	Inherited Disorders of Hemoglobin
SCD	Sickle Cell Disorders
RBC	Red Blood Cells
P. falciparum	*Plasmodium falciparum*
P. malaria	Plasmodium. malariae
P. vivax	Plasmodium. vivax
P. ovale	Plasmoduim. ovale
P. knowlesi	Plasmodium. knowlesi
HbS	Sickle hemoglobin
HbC	hemoglobin C
HbE	hemoglobin E
HbAA	normal genotype (hemoglobin AA genotype)
HbCC	homozygotes for βC (hemoglobin CC genotype)
HbAC	heterozygotes for βC (hemoglobin AC genotype)
HbSS	homozygotes for βS (hemoglobin SS genotype)
HbAS	heterozygotes for βS (hemoglobin AS genotype)
HbSC	heterozygotes for βS and βC (hemoglobin AS genotype)
HbEE	homozygotes for βE (hemoglobin EE genotype)
HbAE	heterozygotes for βE (hemoglobin AE genotype)
G6PD	Deficiency Glucose-6-phosphate dehydrogenase
CM	cerebral malaria
SMA	severe malarial anemia
PfEMP-1	*P. falciparum* erythrocyte membrane protein 1
WHO	World Health Organization
RDTs	Rapid diagnostic Tests
ITNs	insecticide-treated nets
ACT	Artemisinin-based Combination Therapies
IRS	Indoor Residual insecticide Spray
GWA	genome-wide association study

DOI: http://dx.doi.org/10.5772/intechopen.93807

Author details

Edith Christiane Bougouma[1] and Sodiomon Bienvenu Sirima[2*]

1 Institut National de Sante Publique (INSP), Ouagadougou, Burkina Faso

2 Groupe de Recherche Action en Santé (GRAS), Ouagadougou, Burkina Faso

*Address all correspondence to: s.sirima@gras.bf

References

[1] Kariuki SN, Williams TN. Human genetics and malaria resistance. Human Genetics. 2020

[2] WHO. World Malaria Report. https://wwwwhoint/malaria/publications/world-malaria-report-2019/en/2019.

[3] Goncalves BP, Sagara I, Coulibaly M, Wu Y, Assadou MH, Guindo A, et al. Hemoglobin variants shape the distribution of malaria parasites in human populations and their transmission potential. Scientific Reports. 2017;**7**:14267

[4] Roberts DJ, Williams TN. Haemoglobinopathies and resistance to malaria. Redox Report. 2003;**8**:304-310

[5] Weatherall DJ. Genetic variation and susceptibility to infection: The red cell and malaria. British Journal of Haematology. 2008;**141**:276-286

[6] Allison AC. Protection afforded by sickle-cell trait against subtertian malareal infection. British Medical Journal. 1954;**1**:290-294

[7] Hedrick PW. Population genetics of malaria resistance in humans. Heredity (Edinb). 2011;**107**:283-304

[8] Christianson AD, Howson CP, Modell B. Global report on birth defects. White Plains, New York: March of Dimes Birth Defects Foundation; 2006. p. 84

[9] Kwiatkowski DP. How malaria has affected the human genome and what human genetics can teach us about malaria. American Journal of Human Genetics. 2005;**77**:171-192

[10] Kariuki SN, Williams TN. Human genetics and malaria resistance. Human Genetics. 2020;**139**:801-811

[11] Taylor SM, Cerami C, Fairhurst RM. Hemoglobinopathies: Slicing the Gordian knot of plasmodium falciparum malaria pathogenesis. PLoS Pathogens. 2013;**9**:e1003327

[12] Howes R, Battle K, Gething P: Inherited Blood Disorders 2017. https://malariaatlasorg/research.

[13] Singh B, Kim Sung L, Matusop A, Radhakrishnan A, Shamsul SS, Cox-Singh J, et al. A large focus of naturally acquired plasmodium knowlesi infections in human beings. Lancet. 2004;**363**:1017-1024

[14] Mendis K, Sina BJ, Marchesini P, Carter R. The neglected burden of plasmodium vivax malaria. The American Journal of Tropical Medicine and Hygiene. 2001;**64**:97-106

[15] Rogerson SJ, Carter R. Severe vivax malaria: Newly recognised or rediscovered. PLoS Medicine. 2008;**5**: e136

[16] Martinsen ES, Blumberg BJ, Eisen RJ, Schall JJ. Avian hemosporidian parasites from northern California oak woodland and chaparral habitats. Journal of Wildlife Diseases. 2008;**44**:260-268

[17] Lopez C, Saravia C, Gomez A, Hoebeke J, Patarroyo MA. Mechanisms of genetically-based resistance to malaria. Gene. 2010;**467**:1-12

[18] Yuthavong Y, Wilairat P. Protection against malaria by thalassaemia and haemoglobin variants. Parasitology Today. 1993;**9**:241-245

[19] Smith TG, Ayi K, Serghides L, McAllister CD, Kain KC. Innate immunity to malaria caused by plasmodium falciparum. Clinical and Investigative Medicine. 2002;**25**:262-272

[20] Snow RW, Guerra CA, Noor AM, Myint HY, Hay SI. The global distribution of clinical episodes of

plasmodium falciparum malaria. Nature. 2005;**434**:214-217

[21] Hutagalung R, Wilairatana P, Looareesuwan S, Brittenham GM, Aikawa M, Gordeuk VR. Influence of hemoglobin E trait on the severity of falciparum malaria. The Journal of Infectious Diseases. 1999;**179**:283-286

[22] Mackinnon MJ, Mwangi TW, Snow RW, Marsh K, Williams TN. Heritability of malaria in Africa. PLoS Medicine. 2005;**2**:e340

[23] Weatherall DJ, Clegg JB. Genetic variability in response to infection: Malaria and after. Genes and Immunity. 2002;**3**:331-337

[24] Forget BG, Bunn HF. Classification of the disorders of hemoglobin. Cold Spring Harbor Perspectives in Medicine. 2013;**3**:a011684

[25] Sonati Mde F, Costa FF. The genetics of blood disorders: Hereditary hemoglobinopathies. Jornal de Pediatria. 2008;**84**:S40-S51

[26] Bougouma EC, Tiono AB, Ouedraogo A, Soulama I, Diarra A, Yaro JB, et al. Haemoglobin variants and plasmodium falciparum malaria in children under five years of age living in a high and seasonal malaria transmission area of Burkina Faso. Malaria Journal. 2012;**11**:154

[27] Badoum ES, Sermé SS, Yaro JB, Coulibaly SA, Kargougou D, Diarra A, et al. Abnormalities of hemoglobin and Glucose-6-Phosphate-Dehydrogenase deficiency in children with uncomplicated malaria and living in Banfora and Saponé, two different malaria setting of Burkina Faso. International Journal of Tropical Disease & Health. 2019;**37**

[28] Modiano D, Bancone G, Ciminelli BM, Pompei F, Blot I, Simpore J, et al. Haemoglobin S and haemoglobin C: 'quick but costly' versus 'slow but gratis' genetic adaptations to plasmodium falciparum malaria. Human Molecular Genetics. 2008;**17**: 789-799

[29] Luzzatto L. Sickle cell anaemia and malaria. Mediterr J Hematol Infect Dis. 2012;**4**:e2012065

[30] Agarwal A, Guindo A, Cissoko Y, Taylor JG, Coulibaly D, Kone A, et al. Hemoglobin C associated with protection from severe malaria in the Dogon of Mali, a west African population with a low prevalence of hemoglobin S. Blood. 2000;**96**: 2358-2363

[31] Mockenhaupt FP, Ehrhardt S, Cramer JP, Otchwemah RN, Anemana SD, Goltz K, et al. Hemoglobin C and resistance to severe malaria in Ghanaian children. The Journal of Infectious Diseases. 2004; **190**:1006-1009

[32] Goncalves BP, Gupta S, Penman BS. Sickle haemoglobin, haemoglobin C and malaria mortality feedbacks. Malaria Journal. 2016;**15**:26

[33] Taylor SM, Parobek CM, Fairhurst RM. Haemoglobinopathies and the clinical epidemiology of malaria: A systematic review and meta-analysis. The Lancet Infectious Diseases. 2012;**12**: 457-468

[34] Rees DC, Clegg JB, Weatherall DJ. Is hemoglobin instability important in the interaction between hemoglobin E and beta thalassemia? Blood. 1998;**92**: 2141-2146

[35] Bachir D, Galacteros F. Hemoglobin E disease. In: Orphanet Encyclopedia. 2004

[36] Flint J, Harding RM, Boyce AJ, Clegg JB. The population genetics of the haemoglobinopathies. Baillière's Clinical Haematology. 1998;**11**:1-51

[37] Williams TN, Mwangi TW, Wambua S, Peto TE, Weatherall DJ, Gupta S, et al. Negative epistasis between the malaria-protective effects of alpha+−thalassemia and the sickle cell trait. Nature Genetics. 2005;**37**: 1253-1257

[38] Thein SL. The molecular basis of beta-thalassemia. Cold Spring Harbor Perspectives in Medicine. 2013;**3**: a011700

[39] Weatherall DJ, Clegg JB. Inherited haemoglobin disorders: An increasing global health problem. Bulletin of the World Health Organization. 2001;**79**: 704-712

[40] Troye-Blomberg M, Perlmann P, Mincheva Nilsson L, Perlmann H. Immune regulation of protection and pathogenesis in plasmodium falciparum malaria. Parassitologia. 1999;**41**:131-138

[41] Amoah LE, Opong A, Ayanful-Torgby R, Abankwa J, Acquah FK. Prevalence of G6PD deficiency and plasmodium falciparum parasites in asymptomatic school children living in southern Ghana. Malaria Journal. 2016; **15**:388

[42] Beutler E. G6PD: Population genetics and clinical manifestations. Blood Reviews. 1996;**10**:45-52

[43] Ruwende C, Khoo SC, Snow RW, Yates SN, Kwiatkowski D, Gupta S, et al. Natural selection of hemi- and heterozygotes for G6PD deficiency in Africa by resistance to severe malaria. Nature. 1995;**376**:246-249

[44] Modiano D, Luoni G, Sirima BS, Simpore J, Verra F, Konate A, et al. Haemoglobin C protects against clinical Plasmodium falciparum malaria. Nature. 2001;**414**:305-308

[45] Ayi K, Turrini F, Piga A, Arese P. Enhanced phagocytosis of ring-parasitized mutant erythrocytes: A common mechanism that may explain protection against falciparum malaria in sickle trait and beta-thalassemia trait. Blood. 2004;**104**:3364-3371

[46] Williams TN. Human red blood cell polymorphisms and malaria. Current Opinion in Microbiology. 2006;**9**: 388-394

[47] Kar BC, Agrawal KC, Panda A. Sickle haemoglobin, G-6PD deficiency and malaria in western Orissa. The Journal of the Association of Physicians of India. 1990;**38**:555-557

[48] Arya R, Layton DM, Bellingham AJ. Hereditary red cell enzymopathies. Blood Reviews. 1995;**9**:165-175

[49] Angastiniotis M, Modell B. Global epidemiology of hemoglobin disorders. Annals of the New York Academy of Sciences. 1998;**850**:251-269

[50] Mangano VD, Kabore Y, Bougouma EC, Verra F, Sepulveda N, Bisseye C, et al. Novel insights into the protective role of Hemoglobin S and C against plasmodium falciparum Parasitemia. The Journal of Infectious Diseases. 2015;**212**:626-634

[51] Billo MA, Johnson ES, Doumbia SO, Poudiougou B, Sagara I, Diawara SI, et al. Sickle cell trait protects against plasmodium falciparum infection. American Journal of Epidemiology. 2012;**176**(Suppl 7):S175-S185

[52] Weatherall DJ. Recent insights into the population genetics and dynamics of the inherited disorders of hemoglobin. Mediterr J Hematol Infect Dis. 2009;**1**: e200922

[53] Cholera R, Brittain NJ, Gillrie MR, Lopera-Mesa TM, Diakite SA, Arie T, et al. Impaired cytoadherence of plasmodium falciparum-infected erythrocytes containing sickle hemoglobin. Proceedings of the National Academy of Sciences of the

United States of America. 2008;**105**: 991-996

[54] Williams TN, Mwangi TW, Roberts DJ, Alexander ND, Weatherall DJ, Wambua S, et al. An immune basis for malaria protection by the sickle cell trait. PLoS Medicine. 2005;**2**:e128

[55] Pasvol G, Weatherall DJ, Wilson RJ. Cellular mechanism for the protective effect of haemoglobin S against P. falciparum malaria. Nature. 1978;**274**: 701-703

[56] Goheen MM, Wegmuller R, Bah A, Darboe B, Danso E, Affara M, et al. Anemia offers stronger protection than sickle cell trait against the Erythrocytic stage of falciparum Malaria and this protection is reversed by iron supplementation. eBioMedicine. 2016; **14**:123-130

[57] LaMonte G, Philip N, Reardon J, Lacsina JR, Majoros W, Chapman L, et al. Translocation of sickle cell erythrocyte microRNAs into plasmodium falciparum inhibits parasite translation and contributes to malaria resistance. Cell Host & Microbe. 2012; **12**:187-199

[58] Shear HL, Roth EF Jr, Fabry ME, Costantini FD, Pachnis A, Hood A, et al. Transgenic mice expressing human sickle hemoglobin are partially resistant to rodent malaria. Blood. 1993;**81**:222-226

[59] Lelliott PM, McMorran BJ, Foote SJ, Burgio G. The influence of host genetics on erythrocytes and malaria infection: Is there therapeutic potential? Malaria Journal. 2015;**14**:289

[60] Weatherall DJ. The inherited diseases of hemoglobin are an emerging global health burden. Blood. 2010;**115**: 4331-4336

[61] Kilian N, Srismith S, Dittmer M, Ouermi D, Bisseye C, Simpore J, et al. Hemoglobin S and C affect protein export in plasmodium falciparum-infected erythrocytes. Biol Open. 2015; **4**:400-410

[62] Guggenmoos-Holzmann I, Bienzle U, Luzzatto L. Plasmodium falciparum malaria and human red cells. II. Red cell genetic traits and resistance against malaria. International Journal of Epidemiology. 1981;**10**:16-22

[63] Verra F, Simpore J, Warimwe GM, Tetteh KK, Howard T, Osier FH, et al. Haemoglobin C and S role in acquired immunity against plasmodium falciparum malaria. PLoS One. 2007;**2**:e978

[64] Marsh K, Otoo L, Hayes RJ, Carson DC, Greenwood BM. Antibodies to blood stage antigens of plasmodium falciparum in rural Gambians and their relation to protection against infection. Transactions of the Royal Society of Tropical Medicine and Hygiene. 1989; **83**:293-303

[65] Fairhurst RM, Fujioka H, Hayton K, Collins KF, Wellems TE. Aberrant development of plasmodium falciparum in hemoglobin CC red cells: Implications for the malaria protective effect of the homozygous state. Blood. 2003;**101**: 3309-3315

[66] Archer NM, Petersen N, Clark MA, Buckee CO, Childs LM, Duraisingh MT. Resistance to plasmodium falciparum in sickle cell trait erythrocytes is driven by oxygen-dependent growth inhibition. Proceedings of the National Academy of Sciences of the United States of America. 2018;**115**:7350-7355

[67] Gong L, Maiteki-Sebuguzi C, Rosenthal PJ, Hubbard AE, Drakeley CJ, Dorsey G, et al. Evidence for both innate and acquired mechanisms of protection from plasmodium falciparum in children with sickle cell trait. Blood. 2012;**119**:3808-3814

[68] Aidoo M, Terlouw DJ, Kolczak MS, McElroy PD, ter Kuile FO, Kariuki S,

et al. Protective effects of the sickle cell gene against malaria morbidity and mortality. Lancet. 2002;**359**:1311-1312

[69] Guinet F, Diallo DA, Minta D, Dicko A, Sissoko MS, Keita MM, et al. A comparison of the incidence of severe malaria in Malian children with normal and C-trait hemoglobin profiles. Acta Tropica. 1997;**68**:175-182

[70] Malaria Genomic Epidemiology N, Malaria Genomic Epidemiology N. Reappraisal of known malaria resistance loci in a large multicenter study. Nature Genetics. 2014;**46**:1197-1204

[71] Goheen MM, Campino S, Cerami C. The role of the red blood cell in host defence against falciparum malaria: An expanding repertoire of evolutionary alterations. British Journal of Haematology. 2017;**179**:543-556

[72] Willcox M, Bjorkman A, Brohult J, Pehrson PO, Rombo L, Bengtsson E. A case-control study in northern Liberia of plasmodium falciparum malaria in haemoglobin S and beta-thalassaemia traits. Annals of Tropical Medicine and Parasitology. 1983;**77**:239-246

[73] Ackerman H, Usen S, Jallow M, Sisay-Joof F, Pinder M, Kwiatkowski DP. A comparison of case-control and family-based association methods: The example of sickle-cell and malaria. Annals of Human Genetics. 2005;**69**:559-565

[74] Kreuels B, Kreuzberg C, Kobbe R, Ayim-Akonor M, Apiah-Thompson P, Thompson B, et al. Differing effects of HbS and HbC traits on uncomplicated falciparum malaria, anemia, and child growth. Blood. 2010; **115**:4551-4558

[75] Ntoumi F, Mercereau-Puijalon O, Ossari S, Luty A, Reltien J, Georges A, et al. Plasmodium falciparum: Sickle-cell trait is associated with higher prevalence of multiple infections in Gabonese children with asymptomatic infections. Experimental Parasitology. 1997;**87**:39-46

[76] Danquah I, Ziniel P, Eggelte TA, Ehrhardt S, Mockenhaupt FP. Influence of haemoglobins S and C on predominantly asymptomatic plasmodium infections in northern Ghana. Transactions of the Royal Society of Tropical Medicine and Hygiene. 2010;**104**:713-719

[77] Aluoch JR. Higher resistance to plasmodium falciparum infection in patients with homozygous sickle cell disease in western Kenya. Tropical Medicine & International Health. 1997; **2**:568-571

[78] Ringelhann B, Hathorn MK, Jilly P, Grant F, Parniczky G. A new look at the protection of hemoglobin AS and AC genotypes against plasmodium falciparum infection: A census tract approach. American Journal of Human Genetics. 1976;**28**:270-279

[79] Allen SJ, Bennett S, Riley EM, Rowe PA, Jakobsen PH, O'Donnell A, et al. Morbidity from malaria and immune responses to defined plasmodium falciparum antigens in children with sickle cell trait in the Gambia. Transactions of the Royal Society of Tropical Medicine and Hygiene. 1992;**86**:494-498

[80] Thompson GR. Significance of haemoglobins S and C in Ghana. British Medical Journal. 1962;**1**:682-685

[81] Bernstein SC, Bowman JE, Kaptue Noche L. Population studies in Cameroon: Hemoglobin S, glucose-6-phosphate dehydrogenase deficiency and falciparum malaria. Human Heredity. 1980;**30**:251-258

[82] Motulsky AG, Vandepitte J, Fraser GR. Population genetic studies in the Congo. I. Glucose-6-phosphate dehydrogenase deficiency, hemoglobin

S, and malaria. American Journal of Human Genetics. 1966;**18**:514-537

[83] Rihet P, Flori L, Tall F, Traore AS, Fumoux F. Hemoglobin C is associated with reduced plasmodium falciparum parasitemia and low risk of mild malaria attack. Human Molecular Genetics. 2004;**13**:1-6

[84] Lopera-Mesa TM, Doumbia S, Konate D, Anderson JM, Doumbouya M, Keita AS, et al. Effect of red blood cell variants on childhood malaria in Mali: A prospective cohort study. Lancet Haematol. 2015;**2**:e140-e149

[85] Fairhurst RM, Baruch DI, Brittain NJ, Ostera GR, Wallach JS, Hoang HL, et al. Abnormal display of PfEMP-1 on erythrocytes carrying haemoglobin C may protect against malaria. Nature. 2005;**435**:1117-1121

[86] Cyrklaff M, Sanchez CP, Kilian N, Bisseye C, Simpore J, Frischknecht F, et al. Hemoglobins S and C interfere with actin remodeling in plasmodium falciparum-infected erythrocytes. Science. 2011;**334**:1283-1286

[87] Arie T, Fairhurst RM, Brittain NJ, Wellems TE, Dvorak JA. Hemoglobin C modulates the surface topography of plasmodium falciparum-infected erythrocytes. Journal of Structural Biology. 2005;**150**:163-169

[88] Tokumasu F, Fairhurst RM, Ostera GR, Brittain NJ, Hwang J, Wellems TE, et al. Band 3 modifications in plasmodium falciparum-infected AA and CC erythrocytes assayed by autocorrelation analysis using quantum dots. Journal of Cell Science. 2005;**118**: 1091-1098

[89] Gilles HM, Fletcher KA, Hendrickse RG, Lindner R, Reddy S, Allan N. Glucose-6-phosphate-dehydrogenase deficiency, sickling, and malaria in African children in South Western Nigeria. Lancet. 1967;**1**:138-140

[90] May J, Evans JA, Timmann C, Ehmen C, Busch W, Thye T, et al. Hemoglobin variants and disease manifestations in severe falciparum malaria. JAMA. 2007;**297**:2220-2226

[91] Crompton PD, Traore B, Kayentao K, Doumbo S, Ongoiba A, Diakite SA, et al. Sickle cell trait is associated with a delayed onset of malaria: Implications for time-to-event analysis in clinical studies of malaria. The Journal of Infectious Diseases. 2008;**198**:1265-1275

[92] Labie D, Richin C, Pagnier J, Gentilini M, Nagel RL. Hemoglobins S and C in upper Volta. Human Genetics. 1984;**65**:300-302

[93] Storey J, Fleming AF, Cornille-Brogger R, Molineaux L, Matsushima T, Kagan I. Abnormal haemoglobins in the Sudan savanna of Nigeria. IV. Malaria, immunoglobulins and antimalarial antibodies in haemoglobin AC individuals. Annals of Tropical Medicine and Parasitology. 1979;**73**:311-315

[94] Williams TN, Maitland K, Bennett S, Ganczakowski M, Peto TE, Newbold CI, et al. High incidence of malaria in alpha-thalassaemic children. Nature. 1996;**383**:522-525

[95] Fucharoen S, Weatherall DJ. The hemoglobin E thalassemias. Cold Spring Harbor Perspectives in Medicine. 2012;**2**

[96] Chotivanich K, Udomsangpetch R, Pattanapanyasat K, Chierakul W, Simpson J, Looareesuwan S, et al. Hemoglobin E: A balanced polymorphism protective against high parasitemias and thus severe P falciparum malaria. Blood. 2002;**100**: 1172-1176

[97] Fucharoen G, Fucharoen S, Sanchaisuriya K, Sae-Ung N, Suyasunanond U, Sriwilai P, et al. Frequency distribution and Haplotypic heterogeneity of beta(E)-globin gene

among eight minority groups of Northeast Thailand. Human Heredity. 2002;**53**:18-22

[98] Ohashi J, Naka I, Patarapotikul J, Hananantachai H, Brittenham G, Looareesuwan S, et al. Strong linkage disequilibrium of a HbE variant with the (AT)9(T)5 repeat in the BP1 binding site upstream of the beta-globin gene in the Thai population. Journal of Human Genetics. 2005;**50**:7-11

[99] Bunyaratvej A, Butthep P, Sae-Ung N, Fucharoen S, Yuthavong Y. Reduced deformability of thalassemic erythrocytes and erythrocytes with abnormal hemoglobins and relation with susceptibility to plasmodium falciparum invasion. Blood. 1992;**79**: 2460-2463

[100] Oo M, Tin S, Marlar T, O'Sullivan WJ. Genetic red cell disorders and severity of falciparum malaria in Myanmar. Bulletin of the World Health Organization. 1995;**73**:659-665

[101] Kar S, Seth S, Seth PK. Prevalence of malaria in Ao Nagas and its association with G6PD and HbE. Human Biology. 1992;**64**:187-197

[102] Williams TN, Wambua S, Uyoga S, Macharia A, Mwacharo JK, Newton CR, et al. Both heterozygous and homozygous alpha+ thalassemias protect against severe and fatal plasmodium falciparum malaria on the coast of Kenya. Blood. 2005;**106**:368-371

[103] Mockenhaupt FP, Ehrhardt S, Gellert S, Otchwemah RN, Dietz E, Anemana SD, et al. Alpha(+)-thalassemia protects African children from severe malaria. Blood. 2004;**104**: 2003-2006

[104] Allen SJ, O'Donnell A, Alexander ND, Alpers MP, Peto TE, Clegg JB. Weatherall DJ: Alpha+-thalassemia protects children against disease caused by other infections as well as malaria. Proceedings of the National Academy of Sciences of the United States of America. 1997;**94**:14736-14741

[105] Pattanapanyasat K, Yongvanitchit K, Tongtawe P, Tachavanich K, Wanachiwanawin W, Fucharoen S, et al. Impairment of plasmodium falciparum growth in thalassemic red blood cells: Further evidence by using biotin labeling and flow cytometry. Blood. 1999;**93**: 3116-3119

[106] Williams TN, Weatherall DJ, Newbold CI. The membrane characteristics of plasmodium falciparum-infected and -uninfected heterozygous alpha(0)thalassaemic erythrocytes. British Journal of Haematology. 2002;**118**:663-670

[107] Hill AV, Bowden DK, O'Shaughnessy DF, Weatherall DJ, Clegg JB. Beta thalassemia in Melanesia: Association with malaria and characterization of a common variant (IVS-1 nt 5 G——C). Blood. 1988;**72**:9-14

[108] Colbourne MJ, Edington GM. Sickling and malaria in the Gold Coast. British Medical Journal. 1956;**1**:784-786

[109] Cockburn IA, Mackinnon MJ, O'Donnell A, Allen SJ, Moulds JM, Baisor M, et al. A human complement receptor 1 polymorphism that reduces plasmodium falciparum rosetting confers protection against severe malaria. Proceedings of the National Academy of Sciences of the United States of America. 2004;**101**:272-277

[110] Luzzi GA, Merry AH, Newbold CI, Marsh K, Pasvol G. Protection by alpha-thalassaemia against plasmodium falciparum malaria: Modified surface antigen expression rather than impaired growth or cytoadherence. Immunology Letters. 1991;**30**:233-240

[111] Yuthavong Y, Bunyaratvej A, Kamchonwongpaisan S. Increased

susceptibility of malaria-infected variant erythrocytes to the mononuclear phagocyte system. Blood Cells. 1990;**16**: 591-597

[112] Fowkes FJ, Michon P, Pilling L, Ripley RM, Tavul L, Imrie HJ, et al. Host erythrocyte polymorphisms and exposure to plasmodium falciparum in Papua New Guinea. Malaria Journal. 2008;**7**:1

[113] Enevold A, Lusingu JP, Mmbando B, Alifrangis M, Lemnge MM, Bygbjerg IC, et al. Reduced risk of uncomplicated malaria episodes in children with alpha+− thalassemia in northeastern Tanzania. The American Journal of Tropical Medicine and Hygiene. 2008;**78**:714-720

[114] Wambua S, Mwangi TW, Kortok M, Uyoga SM, Macharia AW, Mwacharo JK, et al. The effect of alpha+− thalassaemia on the incidence of malaria and other diseases in children living on the coast of Kenya. PLoS Medicine. 2006;**3**:e158

[115] Veenemans J, Andang'o PE, Mbugi EV, Kraaijenhagen RJ, Mwaniki DL, Mockenhaupt FP, et al. Alpha+ −thalassemia protects against anemia associated with asymptomatic malaria: Evidence from community-based surveys in Tanzania and Kenya. The Journal of Infectious Diseases. 2008;**198**:401-408

[116] Lin E, Tavul L, Michon P, Richards JS, Dabod E, Beeson JG, et al. Minimal association of common red blood cell polymorphisms with plasmodium falciparum infection and uncomplicated malaria in Papua new Guinean school children. The American Journal of Tropical Medicine and Hygiene. 2010;**83**:828-833

[117] Haldane JB. A test for homogeneity of records of familial abnormalities. Annals of Eugenics. 1949;**14**:339-341

[118] Thein SL. Genetic association studies in beta-hemoglobinopathies. Hematology. American Society of Hematology. Education Program. 2013; **2013**:354-361

[119] Kaminsky R, Kruger N, Hempelmann E, Bommer W. Reduced development of plasmodium falciparum in beta-thalassaemic erythrocytes. Zeitschrift für Parasitenkunde. 1986;**72**: 553-556

[120] Senok AC, Li K, Nelson EA, Yu LM, Tian LP, Oppenheimer SJ. Invasion and growth of plasmodium falciparum is inhibited in fractionated thalassaemic erythrocytes. Transactions of the Royal Society of Tropical Medicine and Hygiene. 1997;**91**:138-143

[121] Carlson J, Nash GB. Gabutti V, al-Yaman F, Wahlgren M: Natural protection against severe plasmodium falciparum malaria due to impaired rosette formation. Blood. 1994;**84**: 3909-3914

[122] Penman BS, Pybus OG, Weatherall DJ, Gupta S. Epistatic interactions between genetic disorders of hemoglobin can explain why the sickle-cell gene is uncommon in the Mediterranean. Proceedings of the National Academy of Sciences of the United States of America. 2009;**106**: 21242-21246

[123] Beutler E. The hemolytic effect of primaquine and related compounds: A review. Blood. 1959;**14**:103-139

[124] Beutler E. Glucose-6-phosphate dehydrogenase deficiency: a historical perspective. Blood. 2008;**111**:16-24

[125] Nair PA, Al Khusaiby SM. Kernicterus and G6PD deficiency–a case series from Oman. Journal of Tropical Pediatrics. 2003;**49**:74-77

[126] Ginsburg H, Atamna H, Shalmiev G, Kanaani J, Krugliak M.

Resistance of glucose-6-phosphate dehydrogenase deficiency to malaria: Effects of fava bean hydroxypyrimidine glucosides on plasmodium falciparum growth in culture and on the phagocytosis of infected cells. Parasitology. 1996;**113**(Pt 1):7-18

[127] Cappadoro M, Giribaldi G, O'Brien E, Turrini F, Mannu F, Ulliers D, et al. Early phagocytosis of glucose-6-phosphate dehydrogenase (G6PD)-deficient erythrocytes parasitized by plasmodium falciparum may explain malaria protection in G6PD deficiency. Blood. 1998;**92**:2527-2534

[128] Luzzatto L, Usanga FA, Reddy S. Glucose-6-phosphate dehydrogenase deficient red cells: Resistance to infection by malarial parasites. Science. 1969;**164**:839-842

[129] Usanga EA, Luzzatto L. Adaptation of plasmodium falciparum to glucose 6-phosphate dehydrogenase-deficient host red cells by production of parasite-encoded enzyme. Nature. 1985;**313**: 793-795

[130] Ganczakowski M, Bowden DK, Maitland K, Williams TN, O'Shaughnessy D, Viji J, et al. Thalassaemia in Vanuatu, south-West Pacific: Frequency and haematological phenotypes of young children. British Journal of Haematology. 1995;**89**: 485-495

[131] Tishkoff SA, Varkonyi R, Cahinhinan N, Abbes S, Argyropoulos G, Destro-Bisol G, et al. Haplotype diversity and linkage disequilibrium at human G6PD: Recent origin of alleles that confer malarial resistance. Science. 2001;**293**: 455-462

[132] Sabeti PC, Reich DE, Higgins JM, Levine HZ, Richter DJ, Schaffner SF, et al. Detecting recent positive selection in the human genome from haplotype structure. Nature. 2002;**419**:832-837

[133] Johnson MK, Clark TD, Njama-Meya D, Rosenthal PJ, Parikh S. Impact of the method of G6PD deficiency assessment on genetic association studies of malaria susceptibility. PLoS One. 2009;**4**:e7246

[134] Guindo A, Fairhurst RM, Doumbo OK, Wellems TE, Diallo DA. X-linked G6PD deficiency protects hemizygous males but not heterozygous females against severe malaria. PLoS Medicine. 2007;**4**:e66

[135] Modell B, Darlison M. Global epidemiology of haemoglobin disorders and derived service indicators. Bulletin of the World Health Organization. 2008;**86**:480-487

[136] Weatherall D, Greenwood B, Chee HL, Wasi P: Science and Technology for Disease Control: Past, Present, and Future. In Disease Control Priorities in Developing Countries. Edited by nd, Jamison DT, Breman JG, Measham AR, Alleyne G, Claeson M, Evans DB, Jha P, Mills A, Musgrove P. Washington, DC 2006

[137] Colah R, Gorakshakar A, Phanasgaonkar S, D'Souza E, Nadkarni A, Surve R, et al. Epidemiology of beta-thalassaemia in Western India: Mapping the frequencies and mutations in sub-regions of Maharashtra and Gujarat. British Journal of Haematology. 2010;**149**: 739-747

[138] de Silva S, Fisher CA, Premawardhena A, Lamabadusuriya SP, Peto TE, Perera G, et al. Thalassaemia in Sri Lanka: Implications for the future health burden of Asian populations. Sri Lanka Thalassaemia Study Group. Lancet. 2000;**355**:786-791

[139] O'Riordan S, Hien TT, Miles K, Allen A, Quyen NN, Hung NQ, et al. Large scale screening for haemoglobin disorders in southern Vietnam: Implications for avoidance and

management. British Journal of Haematology. 2010;**150**:359-364

[140] Fattoum S. Evolution of hemoglobinopathy prevention in Africa: Results, problems and prospect. Mediterr J Hematol Infect Dis. 2009;**1**: e2009005

[141] Sachs J, Malaney P. The economic and social burden of malaria. Nature. 2002;**415**:680-685

[142] Opi DH, Swann O, Macharia A, Uyoga S, Band G, Ndila CM, et al. Two complement receptor one alleles have opposing associations with cerebral malaria and interact with alpha(+) thalassaemia. eLife. 2018;**7**

[143] Mangano VD, Clark TG, Auburn S, Campino S, Diakite M, Fry AE, et al. Lack of association of interferon regulatory factor 1 with severe malaria in affected child-parental trio studies across three African populations. PLoS One. 2009;**4**:e4206

[144] Damena D, Denis A, Golassa L, Chimusa ER. Genome-wide association studies of severe P. falciparum malaria susceptibility: Progress, pitfalls and prospects. BMC Medical Genomics. 2019;**12**:120

[145] Manolio TA. Cohort studies and the genetics of complex disease. Nature Genetics. 2009;**41**:5-6