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Plasmodium Species and Drug Resistance

Sintayehu Tsegaye Tseha

Abstract

Malaria is a leading public health problem in tropical and subtropical countries of the world. In 2019, there were an estimated 229 million malaria cases and 409, 000 deaths due malaria in the world. The objective of this chapter is to discuss about the different Plasmodium parasites that cause human malaria. In addition, the chapter discusses about antimalarial drugs resistance. Human malaria is caused by five *Plasmodium* species, namely *P. falciparum*, *P. malariae*, *P. vivax*, *P. ovale* and *P. knowlesi*. In addition to these parasites, malaria in humans may also arise from zoonotic malaria parasites, which includes *P. inui* and *P. cynomolgi*. The plasmodium life cycle involves vertebrate host and a mosquito vector. The malaria parasites differ in their epidemiology, virulence and drug resistance pattern. *P. falciparum* is the deadliest malaria parasite that causes human malaria. *P. falciparum* accounted for nearly all malarial deaths in 2018. One of the major challenges to control malaria is the emergence and spread of antimalarial drug-resistant Plasmodium parasites. The *P. vivax* and *P. falciparum* have already developed resistance against convectional antimalarial drugs such as chloroquine, sulfadoxine-pyrimethamine, and atovaquone. Chloroquine-resistance is connected with mutations in pfcrt. Resistance to Sulfadoxine and pyrimethamine is associated with multiple mutations in pfdhps and pfdhfr genes. In response to the evolution of drug resistance *Plasmodium* parasites, artemisinin-based combination therapies (ACTs) have been used for the treatment of uncomplicated falciparum malaria since the beginning of 21st century. However, artemisinin resistant *P. falciparum* strains have been recently observed in different parts of the world, which indicates the possibility of the spread of artemisinin resistance to all over the world. Therefore, novel antimalarial drugs have to be searched so as to replace the ACTs if Plasmodium parasites develop resistance to ACTs in the future.

Keywords: Malaria, Plasmodium species, antimalarial drug resistance

1. Introduction

Malaria is a leading public health problem in tropical and subtropical countries of the world. The disease is caused by Plasmodium parasites that are transmitted by the bites of infected female Anopheles mosquitoes. In 2019, there were an estimated 229 million malaria cases and 409, 000 deaths due the disease in the world. Children aged under 5 years are the most vulnerable group, which accounted for 67% of all malaria deaths occurred in 2019. Nearly 94% of all malaria cases in 2019 occurred in Africa [1].

In addition to its health burden, malaria has also placed a heavy economic burden in Africa. Since 2000, the average annual cost of case management alone has been estimated nearly USD 300 million in Africa [2]. One of the main challenges of controlling malaria is the evolution of drug resistant strains of Plasmodium species against available antimalarial drugs [3–5]. In this chapter, antimalarial drugs resistance has been discussed. Furthermore, the life cycle, clinical features and chemotherapy of the different species of human malaria parasites have been discussed.

2. Plasmodium species causing human malaria

2.1 Diversity of human malaria parasites

Malaria is caused by protozoan parasites that belongs to the genus Plasmodium. There are over 200 plasmodium species that have been proven to cause malaria. But human malaria is caused by only by five Plasmodium species, namely: - *Plasmodium falciparum* (*P. falciparum*); *Plasmodium vivax* (*P. vivax*); *Plasmodium malariae* (*P. malariae*), *Plasmodium ovale* (*P. ovale*) and *Plasmodium knowlesi* (*P. knowlesi*) [6]. While the first four plasmodium species (*P. falciparum*, *P. vivax*, *P. malariae* and *P. ovale*) naturally cause malaria only in humans, *P. knowlesi* causes zoonotic malaria in South East Asia, that is naturally maintained in macaque monkeys. The malaria parasites differ in their epidemiology, virulence and drug resistance pattern. Of the five human malaria parasites, *P. vivax* and *P. falciparum* pose the greatest threat. *P. falciparum* is the most dangerous malaria parasite that is responsible for high morbidity and mortality [7, 8]. *P. falciparum* is the most prevalent human malaria parasite in Africa, that accounted for 99.7% of the estimated malaria cases in 2018 [9].

Plasmodium parasites belong to the order *Haemosporidia* [10]. The different Plasmodium species have different host range. For example, the host range of *P. relictum* is so broad, that can infect more than 100 different species of birds, that belong to different orders and families [11]. Whereas, the host range of *P. falciparum* is so narrow that only infects humans [12]. In contrast to mammalian malaria parasites, that are only transmitted by are mosquitoes of the genus *Anopheles*, *Plasmodium* species that infect birds are transmitted by a wide variety of mosquitoes including *Culex* and *Aedes* [13].

2.2 The life cycle of malaria parasites

The life cycle of human malaria parasites is generally the same [14]. The plasmodium life cycle involves two hosts (has two parts). In the first part, the parasite infects a vertebrate host such as human being and in the second part, the plasmodium parasite infects the mosquito vector.

The Plasmodium life cycle starts when sporozoites enter the blood of the vertebrate host following a bite by the mosquito vector [15]. Then, the sporozoites rapidly move to the liver and invade hepatocytes where they multiply asexually by a process called schizogony (exo-erythrocytic schizogony) and produce merozoites [16, 17]. The merozoites are released back into the blood and infect erythrocytes [18]. In only *P. ovale* and *P. vivax* infection, some of the merozoites in the liver may differentiate into a dormant stage (hypnozoite), which may recure again (cause relapse by invading blood stream) after some time in the future unless treated with primaquine. In the infected erythrocyte, each merozoite multiplies by schizogony (erythrocytic schizogony) to produce between 8 and 64 new merozoites, depending on the species of the plasmodium parasite [19]. The newly produced merozoites are

released back to the blood, and continues its intraerythrocytic propagation cycle every 72 (*P. malariae*) hours; every 24 hours (*P. knowlesi*), and every 48 hours (*P. falciparum*, *P. ovale*, *P. vivax*). Some of the merozoites differentiate into male and female gametocytes for sexual reproduction [20, 21].

The second part of the plasmodium life cycle starts when the insect vector bites infected vertebrate host (such as infected person) and the insect ingests the blood containing gametocytes. The gametocyte completes its development in the lumen of the mosquito midgut and the male and the female gametes fuse to produce a zygote [22], which is the only stage with diploid chromosome (genome) [23].

Following fertilization, the zygote undergoes meiosis and differentiates into ookinete (motile form) that has a haploid genome [24]. Then ookinete penetrates the wall of the midgut of the mosquito and forms an oocyst [25]. In the oocyst, mitosis take place repeatedly, and numerous sporozoites are produced by sporogony [26, 27]. When the oocyst matures, it ruptures and releases sporozoites into the haemolymph. Then, the sporozoites migrate to the salivary glands, where they mature and acquire the capacity to infect vertebrate host cells [28]. This cycle (second part of the plasmodium parasite life cycle (from gametocytes to sporozoites) takes about 10–18 days.

2.3 *Plasmodium falciparum*

P. falciparum is the deadliest Plasmodium species that causes human malaria [29]. According to the World Health Organization (WHO), *P. falciparum* accounted for nearly all malarial deaths (99.7%) in 2018, which caused an estimated 405,000 deaths [9]. With the exception of Europe, *P. falciparum* is found in all continents of the world. Before 1970s, *P. falciparum* malaria was common in Europe. But interventions which includes appropriate case management, insecticide spraying, and environmental engineering since the early 20th century resulted in complete eradication in the 1970s [30]. Unlike other malaria endemic countries in which non-falciparum malaria predominates, over 75% of malaria cases were due to *P. falciparum* in Sub-Saharan Africa [31], where 94% of malaria deaths occur [9].

The average incubation period of *P. falciparum* malaria is 11 days (ranging from 9 to 30 days) [32], which is the shortest among Plasmodium species. The sign and symptoms of uncomplicated malaria include fever, headache, nausea, vomiting, and diarrhea. If left untreated, *P. falciparum* malaria usually develops to severe malaria, which may bring about death. Children with severe malaria frequently develop severe anemia and or respiratory distress [33]. Multi-organ failure is common in adults with severe malaria. Acute respiratory distress occurs in 5–25% of adults and up to 29% of pregnant women [34].

The WHO recommends Artemisinin-based combination therapies (ACTs) as first-line treatment for uncomplicated *P. falciparum* malaria. The ACTs that are recommended by the WHO for the treatment of uncomplicated *P. falciparum* malaria include Artemether/lumefantrine, dihydroartemisinin/piperaquine, artesunate/amodiaquine, artesunate/mefloquine, and artesunate/sulfadoxine-pyrimethamine [35]. The choice of ACT that be used in different parts of the world is different, that depends on the level of resistance to the constituents in the ACT. In a condition when first line therapy fails, the following alternative antimalarial drugs can be used as second-line treatment: - artesunate plus tetracycline or doxycycline or clindamycin, and quinine plus tetracycline or doxycycline or clindamycin which is given for seven days.

The recommended first-line treatment of uncomplicated *P. falciparum* malaria in pregnant women during the first trimester is quinine plus clindamycin for seven days [35]. The second line therapy for the treatment of uncomplicated *P. falciparum*

malaria in pregnant women is artesunate plus clindamycin for 7 days. Atovaquone/proguanil, or artemether/lumefantrine or quinine plus doxycycline or clindamycin are recommended for treatment of malaria in travelers returning to non-malaria endemic countries [35]. The recommended treatment for severe *P. falciparum* malaria in adults is intravenous (IV) or intramuscular (IM) artesunate. Quinine is also recommended as an alternative treatment of severe *P. falciparum* malaria if parenteral artesunate is not available [35]. Whereas, artesunate (IV or IM), quinine (IV or IM), and artemether IM are recommended for treatment of severe *P. falciparum* malaria in children, especially in malaria-endemic areas of Africa [35].

2.4 *Plasmodium vivax*

P. vivax is the second important malaria parasite after *P. falciparum*, that causes significant morbidity. The parasite can cause severe disease and even death that is usually associated with splenomegaly [36, 37]. One of the important features that distinguishes *P. vivax* from *P. falciparum* is the occurrence of dormant stage in the liver (hypnozoites) that can be reactivated later in life.

The burden of *P. vivax* malaria differ from one region of the world to the other, which is mainly seen in central Asia (82%), the Americas (6%), Southeast Asia (9%), some parts of Africa (3%) [38, 39]. *P. vivax* has wider distribution than *P. falciparum*, which is associated with the dormant stage of *P. vivax* and the ability of *P. vivax* to survive and reproduce in the mosquito vector at lower temperatures and higher altitudes. In Africa, *P. vivax* is limited to parts of horn of Africa and Madagascar, unlike the other parts of Africa that is not affected by *P. vivax* infection due to the deficiency of Duffy antigen (which serves as receptor for the parasite) in the population [40]. It has been suggested that *P. vivax* originated in Africa. This is based on the fact that gorillas and wild chimpanzees in central Africa are naturally infected with plasmodium parasite that are closely related to the *P. vivax* that causes human malaria [41].

Usually, *P. vivax* causes mild disease, that causes fever, cough, abdominal pain and diarrhea. However, the parasite may cause serious conditions like respiratory distress. In pregnant women, *P. vivax* infection brings about low birth weight. In rare cases, complications might arise from *P. vivax* infection, which includes acute kidney failure, neurological abnormalities, hypoglycemia and low blood pressures, jaundice and coagulation defects [42]. Chloroquine is the drug of choice for the treatment of malaria that is caused by *P. vivax* [43]. However, in areas where the parasite has developed resistance for Chloroquine, such as Papua New Guinea, Korea, and India where chloroquine resistance has grown up to 20% resistance [44], chloroquine has been replaced by other drugs.

2.5 *Plasmodium ovale*

P. ovale is one of the five human malaria parasites. Unlike *P. falciparum* and *P. vivax*, *P. ovale* accounts very small proportion (5%) of the disease [45]. The species *P. ovale* is consisted of two subspecies, *P. ovale curtisi* and *P. ovale walikeri* [46]. *P. ovale* is mainly found in Islands in western pacific and Sub-Saharan Africa [45, 47]. But the parasite also exists in Thailand [48], Vietnam [49] Guinea [50], Bangladesh [51], Cambodia [52] India, [53] and Myanmar [54]. The incubation period of *P. ovale* ranges from 12 to 20 days. The parasite causes very mild disease and it is less dangerous than *P. falciparum*. Like *P. vivax*, *P. ovale* has a dormant stage in the liver (hypnozoites) that can be reactivated later in life [45]. Chloroquine is the drug of choice for the treatment of malaria that is caused by *P. ovale* [55].

2.6 Plasmodium malariae

P. malariae is one of the five human malaria parasites. It causes mild disease, which is therefore called benign malaria. *P. malariae* is found in the Amazon Basin of South America, sub-Saharan Africa, much of southeast Asia, Indonesia, and on many of the islands of the western Pacific [56]. The parasite causes a chronic infection that may sometimes last for a lifetime. Some of the major defining features of *P. malariae* include its longer (72-hour developmental cycle) (quartan periodicity) (compared with the 48-hour cycle of *P. vivax* and *P. falciparum*) and lower parasitemia compared to those in patients infected with *P. falciparum* or *P. vivax* [57, 58]. The signs and symptoms of *P. malariae* malaria include fever, chills and nausea and edema and the nephrotic syndrome has been documented with some *P. malariae* infections [57]. Like that of *P. vivax* and *P. ovale*, Chloroquine is also highly effective against *P. malariae* malaria [55].

2.7 Plasmodium knowlesi

P. knowlesi is the only human malaria parasite that can naturally cause malaria in humans and other non-human primates (NHP) such as macaque monkeys [59, 60]. *P. knowlesi* is closely related to *P. vivax* and other Plasmodium species that infect non-human primates [61].

The parasite exists in South East Asia [62]. *P. knowlesi* rarely reported from areas outside South East Asia because its vector (the mosquitoes it infects: *Anopheles hacker* and *Anopheles latens*) are restricted to South East Asia [63, 64]. *P. knowlesi* has three subspecies which includes *P. knowlesi edesoni*, *P. knowlesi sintoni*, and *P. knowlesi arimai* [65, 66].

In humans, the parasite can cause both severe and uncomplicated malaria [67]. Uncomplicated *P. knowlesi* malaria is manifested by fever, chills, headaches, joint pain, malaise, abdominal pain, diarrhea, and loss of appetite [67]. In contrast to the other human malarias, *P. knowlesi* malaria has daily or quotidian malaria (a fever that spikes every 24 hours) [67, 68]. Like that of *P. vivax*, *P. malariae* and *P. ovale*, Chloroquine is also highly effective against *P. knowlesi* malaria [55].

2.8 Zoonotic malaria parasites

In addition to *P. knowlesi*, other Plasmodium species have also reported to cause zoonotic malaria [69, 70]. Macaques have been reported to be reservoir of six Plasmodium species, namely *P. knowlesi*, *P. inui*, *P. cynomolgi*, *P. coatneyi*, *P. fieldi* and *P. simiovale* in Sarawak in Malaysian Borneo [71]. From these six Plasmodium species, *P. cynomolgi* has been shown to naturally cause human infection [72], and *P. inui* can cause infection in experimental condition [73], suggesting that these species might become the next Plasmodium species that may affect human health in the future.

Zoonotic malaria has also been reported from other parts of the world. Zoonotic malaria that are caused by *P. simium* [74] and *P. brasilianum* [75], that naturally infect platyrrhine monkeys have been reported in South America. *P. simium* and *P. brasilianum* are closely related with *P. vivax* and *P. malariae*, respectively [76].

3. Antimalarial drugs resistance

There are five groups of antimalarial drugs that are currently used for treatment of human malaria, that are classified on basis of their structure and action [77, 78].

The five classes include: - (1). antifolates (pyrimethamine, proguanil, sulfadoxine), (2). 4-aminoquinolines (like chloroquine, amodiaquine, hydroxychloroquine, and aryl amino alcohols (such as quinine, mefloquine) (3). Endoperoxides (like artemisinin and its derivatives), (4) naphthoquinones (like atovaquone), and (5). 8-aminoquinolines (primaquine, tafenoquine). 4- aminoquinolines, antifolates, naphthoquinones and aryl amino alcohols cause inhibition by detoxification of haem, pyrimidine biosynthesis and mitochondrial cytochrome b involved in oxidoreduction, respectively. Whereas endoperoxides, such as artemisinin, act on multiple cellular processes involving reactive oxygen species in Plasmodium cells [78].

One of the major challenges to control malaria is the emergence and spread of antimalarial drug-resistant plasmodium parasites [3–5]. The most important plasmodium parasites (*P. vivax* and *P. falciparum*) have already developed resistance against conventional antimalarial drugs such as chloroquine, sulfadoxine-pyrimethamine, atovaquone [79–85]. In response to the evolution of drug resistance strains of *P. falciparum* malaria, since the mid-2000s, artemisinin-based combination therapies (ACTs) constitute the standard of care for uncomplicated falciparum malaria and are increasingly also taken into consideration for the treatment of non-falciparum malaria (*P. ovale*, *P. vivax*, *P. knowlesi* and *P. malariae*) [35].

The artemisinin and its derivatives in ACTs confer rapid and potent effectiveness, whereas their partner drugs are longer-lived antimalarial (such as lumefantrine, mefloquine, piperazine (PPQ), amodiaquine or sulfadoxine-pyrimethamine). The reason for use of ACTs is the fact that the artemisinin and its derivatives rapidly eliminate the majority of the parasites within days by mechanisms that are distinct from those of the partner drug, which eliminates residual parasites over weeks, so that parasites that may develop resistance to the artemisinin drug would still be eliminated by the partner drug [86].

There is also widespread resistance of *P. vivax* to chloroquine and sulfadoxine-pyrimethamine [84, 85, 87, 88]. Since the early 1990s chloroquine-resistant *P. vivax* (CRPV) has been reported from different parts of the world, mostly from Papua New Guinea, the Solomon Islands and Indonesia, Burma (Myanmar), India, Vietnam, Turkey, and Central and South America [83]. In Africa, chloroquine resistance started in late 1970s, and treatment failure became alarmingly high until the introduction of ACT in 2005 [89, 90]. Now, the recommended drugs for the treatment of CRPV malaria by the U.S. Centers for Disease Control and Prevention (CDC) succeeded by primaquine include; quinine sulfate plus either doxycycline or tetracycline; atovaquone-proguanil; and mefloquine [91]. ACTs has been demonstrated to be effective for both chloroquine-resistant and chloroquine-sensitive strains of *P. vivax* malaria. Therefore, ACTs can be now used to treat malaria caused by *P. vivax* [92–94]. So far, there are no reports of *P. vivax* resistance to artemisinins. The main drawback of using ACTs for the treatment of *P. vivax* malaria with ACTs is that the dormant liver-stage (hypnozoites) are not targeted by the ACTs, and, therefore, primaquine is necessarily required in combination with ACTs to prevent relapse.

Chloroquine targets the polymerization of free haem (the toxic substance for the parasite) within the food vacuole of the parasite. The drug disrupts haemozoin formation so that the parasite dies by the effect of the poisonous haem. The mechanism of chloroquine resistance is drug efflux via the *P. falciparum* chloroquine-resistance transporter (encoded by *pfcr1*) located at the food vacuole. Chloroquine-resistance was connected with mutations in *pfcr1* [95–97].

Sulfadoxine and pyrimethamine inhibit two enzymes of *P. falciparum* that involve in the folate pathway, that are dihydropteroate synthase (PfDhps) and dihydrofolate reductase (PfDhfr), respectively. Resistance to these antimalarials

arises from multiple mutations in *pfdhps* and *pfdhfr* genes [98–101]. In Ethiopia, *P. falciparum* resistance to Sulfadoxine-pyrimethamine (SP) had led to replacement of SP with ACT, which is composed of consisted of artemether and lumefantrine (Coartem) in 2004 [102]. However, artemisinin resistance in *P. falciparum* has emerged in different parts of the world, especially in Southeast Asia and Africa [103–106], which indicates the possibility of the spread of artemisinin resistance to all over the world.

4. Conclusions

Malaria remains the major public health problem in tropical and subtropical countries of the world. Human malaria is caused by five *Plasmodium* species, namely *P. falciparum*, *P. malariae*, *P. vivax*, *P. ovale* and *P. knowlesi*. In addition to these parasites, malaria in humans can sometimes arise from zoonotic malaria parasites, which includes *P. inui*, *P. cynomolgi*, *P. coatneyi*, *P. fieldi*, *P. simiovale*, *P. simium* and *P. brasilianum*. The plasmodium life cycle involves two hosts (has two parts). In the first part, the parasite infects a vertebrate host such as human being and in the second part, the plasmodium parasites infect the mosquito vector. The malaria parasites differ in their epidemiology, virulence and drug resistance pattern. *P. falciparum* is the deadliest *Plasmodium* species that causes human malaria. *P. falciparum* accounted for nearly all malarial deaths (99.7%) in 2018. One of the major challenges to control malaria is the emergence and spread of antimalarial drug-resistant plasmodium parasites. The most important *Plasmodium* parasites (*P. vivax* and *P. falciparum*) have already developed resistance against convectional antimalarial drugs such as chloroquine, sulfadoxine-pyrimethamine, atovaquone. In response to the evolution of drug resistance *Plasmodium* parasites, ACTs have been used as first line therapy for treatment of uncomplicated falciparum malaria since the beginning of 21th century. However, artemisinin resistant *P. falciparum* strains have been recently observed in different parts of the world, which indicates the possibility of the spread of artemisinin resistance to all over the world. Therefore, novel antimalarial drugs have to be searched so as to replace the ACTs if *Plasmodium* parasites develop resistance to ACTs in the future.

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Conflict of interest

The author declares no conflict of interest.

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