Malaria: Transmission, Diagnosis and Treatment: A Review

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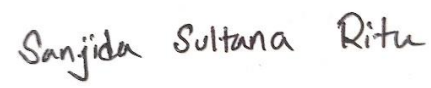

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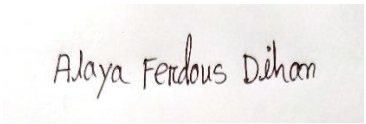

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Abstract

Malaria is an ancient vector-borne human diseases that is caused by the plasmodium parasite. *Anopheles* mosquitoes are the vector that contains the plasmodium parasite sporozoites. There are 5 most common Plasmodium parasite species that can cause human infection. Those are *Plasmodium vivax, Plasmodium ovale, Plasmodium malariae, Plasmodium falciparum,* and *Plasmodium knowlesi*. In 2019, there were about 229 million cases of malaria worldwide. Africa is the most affected country of malaria. The spread and distribution of vector-borne diseases are greatly affected by environmental and climatic factors. Climatic factors especially temperature, humidity and rainfall, affect the ability of malaria parasites and Anopheles vectors to exist long enough to spread disease. An epidemic of malaria occurs when climate and other conditions suddenly favor the spread of malaria in areas where people have little or no immunity to malaria. In China, a sweet sagewort plant known as Qinghai (Latin Artemisia annua) was used to treat malaria as early as the 2nd century BC. Later, Different antimalarial drugs like quinolines, antifolates, and Artemisinin-combination therapies (ACTs) are used to treat malaria. Recently, The World Health Organization (WHO) recommends that children in Sub-Saharan Africa and other places with moderate to high P. falciparum malaria transmission get the RTS,S/AS01 (RTS,S) malaria vaccine. The proposal is based on the outcomes of a trial program that has served over 800,000 children in Ghana, Kenya, and Malawi since 2019.

Keywords: Malaria, *Anopheles*, plasmodium, temperature, transmission, antimalarial drugs, malaria vaccine

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List of Acronyms

ACT	Artimisinin combination therapy
WHO	World health organization
RDT	Rapid Diagnostic Test
ELISA	Enzyme-Linked Immunosorbent Assay
PCR	Polymerase Chain Reaction
CNS	Central Nervous System
CDC	Centers for Disease control and Prevention
IFA	Indirect Immunofluorescence Assay
DDT	Dichlorodiphenyltrichloroethane

Chapter 1

Introduction

Malaria is a very common vector-borne infectious disease that is caused by parasites of the genus *Plasmodium.* A mosquito named *Anopheles* transmitted this disease to humans through a bite of an infected female mosquito of the species. Malaria is the most common disease in Africa and some Asian countries with the highest number of indigenous cases. In 2019, there were about 229 million cases of malaria worldwide. Africa is the most affected country of malaria (WHO 2021). According to the World Health Organization, children who are under 5 years old are mostly affected and died.

There are 5 most common Plasmodium parasite species that can cause human infection. Those are *Plasmodium vivax, Plasmodium ovale, Plasmodium malariae, Plasmodium falciparum,* and *Plasmodium knowlesi*. Among them 2 species of *Plasmodium falciparum* and *Plasmodium vivax* – can cause the greatest threat (95% of infections) to human. Plasmodium *vivax* is responsible for causing 75% of infection (WHO 2021), since it is distributed widely throughout the tropics, subtropics, and temperate zones (Snow et al., 2015). On the other hand, other Plasmodium species are not widely distributed. *Plasmodium falciparum* is generally confined to tropical areas and *Plasmodium malariae* is sporadically distributed. *Plasmodium ovale* is mainly confined to West Central Africa and some South Pacific islands.

Climate and season change have various effects on seasonal transmission of vector-borne diseases, health, even in distribution of pathogens (Vasseur et al., 2014). In some cases, increasing temperatures might affect diseases, carried by mosquitoes such as Dengue fever, malaria may be more worrisome. Meanwhile, other environmental changes such as

deforestation, changes in precipitation can also affect the distribution of mosquitos, their life cycle and therefore affect the spread of mosquito-borne diseases.

In this review, we have explored the route of malaria transmission, different diagnosis methods and its treatment.

Chapter 2

Malaria transmission and life cycle

The spread and distribution of vector-borne diseases are greatly affected by environmental and climatic factors. Climatic factors especially temperature, humidity and rainfall, affect the ability of malaria parasites and Anopheles vectors to exist long enough to spread disease (Vasseur et al., 2014). Mosquito bite is root cause of malaria, so, the mosquito is responsible for infection? No. mosquitos do not cause malaria but just spread it. The actual causative agent of malaria is the protozoan called plasmodium. *Plasmodium vivax, Plasmodium malaria, Plasmodium falciparum* are a few species of plasmodium that cause malaria in human. Among this, *plasmodium falciparum* is the most violent, and can also be life threatening.

The protozoan Plasmodium lives as a parasite in the female Anopheles mosquito. From there, it gets transmitted to humans with a mosquito bite. Hence, it is the infected mosquito bite which can cause malaria and not the insect itself. Thus, the complete life cycle of malarial parasitic protozoa involves two hosts. One is female Anopheles mosquito and another one is the human host.

The complete life cycle of malaria parasite begins when an infected mosquito bites a human. This helps transfer the malarial parasite in the human body, female anopheles mosquito secretes its saliva into human bloodstream to prevent blood coagulation. It is with the secretion; parasite enters the human body. The disease-causing plasmodium can get transferred from mosquitoes to humans in the form of sporozoites. Sporozoites is a stage in the lifecycle of Plasmodium. At this particular stage, they are capable to cause an infection in the human body. When sporozoites enter the bloodstream of human, they eventually reach their target that is liver. Then, they attack the liver cells and stay inside them for quite a long time. Then, sporozoites reproduce asexually, in order to increase their number. After that, they release themselves back

into the bloodstream by bursting out the liver cells. The form in which they are released into the bloodstream is known as merozoites. These merozoites now target the red blood cells, the RBCs. Now, in the RBCs, the merozoites develop into a ring like structure called Trophozoite. This form can have two fates. Most of the trophozoites begin with asexual cycle to give rise to new merozoites and this occurs exponentially. The number of merozoites produced is too large for the RBCs to contain. Hence, they released out with the bursting of the RBCs. The released merozoites can now attack new RBCs and keep increasing their population exponentially. The other fate of trophozoite is entering a sexual cycle. With this, the trophozoites give rise to two different gametocytes. Gametocytes are like the germ cells of plasmodium. When a non-infected mosquito approaches the infected human for a blood meal, then these gametocytes quickly pass into the mosquito's body with the sucked blood. Now, in mosquito's body, both male and female gametocytes, which are like the germ cells fuse to form a zygote. That is, the fertilization of these gametocytes results in the formation and development of zygote in the mosquito's gut. From here, a structure called oocyst is developed which crosses the gut wall and reaches the salivary gland (Hillyer et al., 2007). Here, oocyst releases several sporozoites which are the matured infective form of the parasite ready to infect new healthy individual with the next blood meal. So, when the female anopheles mosquito bites a healthy person, the sporozoites get transmitted, thus repeating the complete cycle and causing the infection. Every person infected with malaria has the parasite undergoing exactly the same morphological changes during its life cycle and human-parasite interactions.

Malaria epidemics can occur when climate and other conditions suddenly favor transmission in areas where people have little or no immunity to malaria. They can also occur when people with low immunity move into areas with intense malaria transmission, for instance to find work, or as refugees. Human immunity is another important factor, especially among adults in areas of moderate or intense transmission conditions.

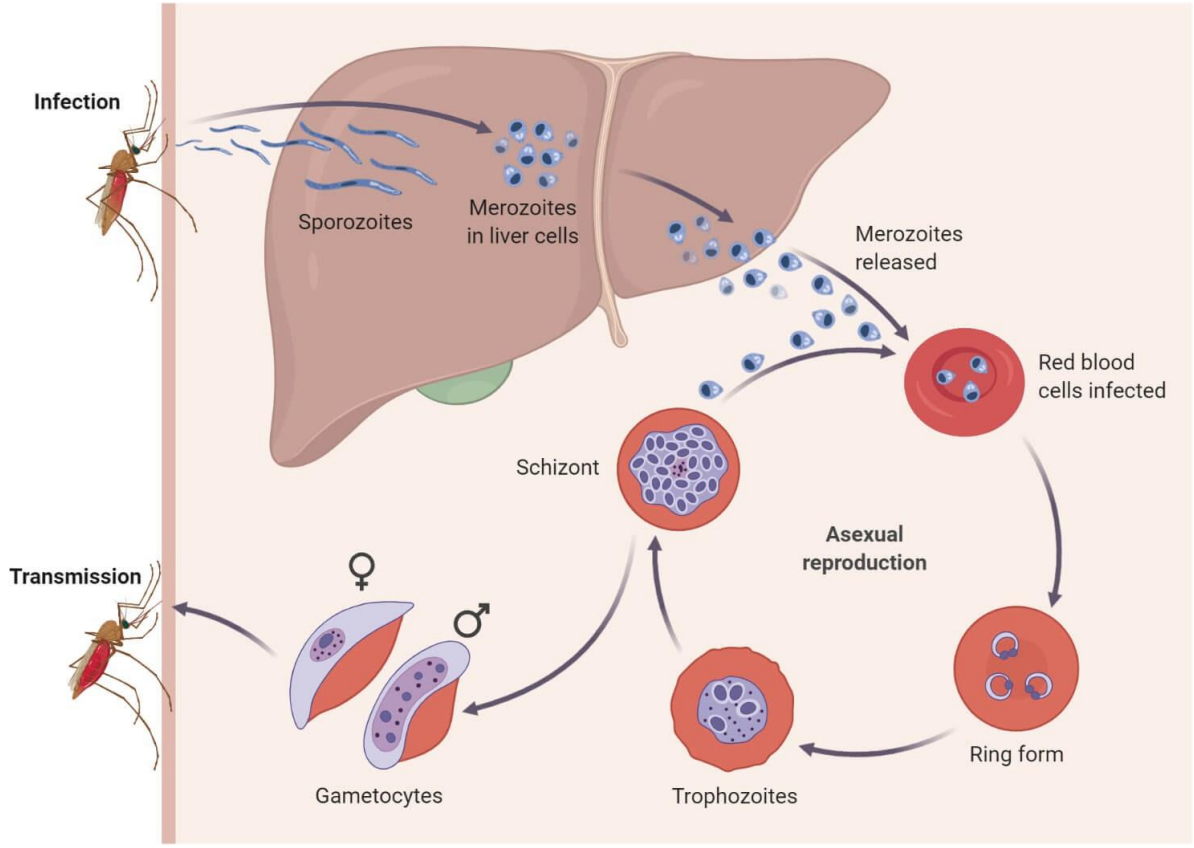

Figure1: This figure is graphically representing the transmission and life cycle of malaria (Neupane et al., 2020)

Chapter 3

People who are at risk

Generally, Malaria occurs in tropical and subtropical areas of the world, mostly in Africa, Central and South America, Asia, and the Indo-Pacific region. When it rains a lot in the tropics, mosquitoes can breed more in numbers as *Anopheles* mosquitoes lay their eggs in the water, from which the larvae hatch and eventually hatch as adult mosquitoes (WHO 2021) and the area is likely suitable for transmission of malaria. Hilly areas are also areas of greatest risk of malaria transmission. Migration of people from populated area to rural hilly areas for agriculture, plantation work is believed to pose a significant risk of spreading malaria.

However, a certain population group has the highest risk of malaria infection. In case of high transmission areas, children under the age of 5 and women who are pregnant with their first child are more susceptible to infection (WHO 2021). On the other hand, in low transmission areas, people of all ages are at risk, because of weakened immunity. Even Refugees are also at very risk due to poor living conditions and weak immunity system such as Rohinga population in Bangladesh. Even, International travelers from non-endemic areas are also at risk as they lack immunity (WHO 2021).

Chapter 4

Effects of climate change on malaria transmission

Climate change act as an important factor for affecting vector population dynamics and therefore also an important factor for increasing the risk of vector-borne diseases. Malaria is one of the most common vectors borne diseases that will be severely affected by global warming and climate change (Houghton et al., 1997). In case of malaria transmission Climate change may play a significant role by changing in the ecology and human behavior, vector Anopheles mosquitoes and Plasmodium parasite (Martens et al., 1997). Probably the vector population would be affected most. (Parham et al., 2010). However, the dynamics of malaria transmission are strongly affected by various environmental conditions such as temperature, precipitation, wind speed, humidity, deforestation, altitude etc. (Vasseur et al., 2014).

4.1 Temperature

It is predicted that by 2100, the average temperature of the earth might increase by 2.6 to 4.8°C. (Hanafi-Bojd et al., 2019). The increase in temperature has an impact on the abundance of the vector, because temperature, as a physiological factor, affects the shortening of the life cycle of the Anopheles mosquito, from egg to adult by affecting the development rate of larvae and the probability of adult survival. (Martens et al., 1998; Martens P et al., 2000 & Parham et al., 2010). Then, as its annual generation number increases, the vector population will increase. Moreover, the growth and development of malaria parasites in female mosquitoes will accelerate due to global warming and increase the blood pressure of humans, thus increasing human-mosquito contact. All these factor combination will increase the likelihood of being bitten by malaria vectors. Therefore, in some parts of the world, people believe that the burden of malaria will increase (Blanford et al., 2013; Siraj et al., 2014). Recent evidence shows that

the incidence of malaria is increasing in colder parts of the world due to global warming (Murdock et al., 2016).

4.2 Precipitation

Precipitation is another factor that mainly affects the vector population itself. It is important to consider the impact of climate change on rainfall, because it can affect the formation and existence of mosquito breeding sites (Afrane et al., 2012). Since, the immature stages of the mosquito life cycles are aquatic and rainfall can lead to increased availability of breeding sites, which act as a strong driving force for sufficient vector population (Parham et al., 2010).

4.3 Deforestation

Deforestation is a constant problem in developing countries that creates suitable conditions for the spread of vectors and the spread of infectious diseases. Deforestation has changed the dynamics of ecosystems and creates new breeding habitats for disease vectors (such as mosquitoes and fleas) by recreating the boundaries of existing ecosystems. These boundaries are usually places where humans come into contact with forest pathogens. (Gottwalt et al., 2013).Developing countries continue to deforest for agriculture to meet the needs of their growing population. Due to increased vector contact with forest edges and the reduction of biodiversity in the area, people living in or near these fragmented forests are at a much higher risk of developing zoonotic diseases. Therefore, changes in forest cover may have a wide-ranging and multi-factorial impact on the ecology of mosquito vectors and their spreading pathogens, and determine the geographic distribution and burden of some of the most common and debilitating human infectious diseases worldwide, such as malaria And dengue fever (Macdonald et al., 2019). As the global deforestation rate is expected to continue to rise in the foreseeable future, its impact on vector-borne infectious diseases is also expected to increase, and the scope and spread of malaria will continue to expand, if no appropriate step is taken.

Chapter 5

Malaria diagnosis

For a laboratory examination of malaria, firstly need to know about clinical results. Clinical diagnosis based on patient symptoms and physical results in the test. Initially, the most common symptoms of malaria are fever, chills, headache, sweating, muscle pain, and vomiting. The results of the physical tests are also mostly unspecific such as increased body temperature, fatigue etc. There are direct (Microscopic analysis, Rapid diagnostic tests, Molecular tests) and indirect (Indirect immunofluorescence, ELISA) test methods for malaria diagnostic.

5.1 Light Microscopic analysis

A high standard, light microscopy is performed to identify accurately Plasmodium parasites with sufficient rapidity. It remains the Operational Gold standard both in the control and disposal configuration. To performing the test blood smear is prepared the sample is stained with Giemsa stain to give the parasite a unique appearance. Due to the lack of suitable staining materials and well-trained technicians, this method is not available in many regions of sub-Saharan Africa (Kachur et al., 1995). However, it depends on the quality of the reagents, the microscope and the experience of the laboratory technicians.

5.2 Rapid diagnostic tests

Rapid Diagnostic Tests (RDT) is immunochromatographic tests (most often use a dipstick or cassette format,) used for the detection of antigens in the blood are to prove the presence of parasite antigens. To perform this test any electrical equipment's are not needed or no special skills are required. RDTs can provide results within 2-15 minutes. It can be used as a useful alternative of microscopy analysis methods, where reliable microscopic diagnosis is not available. A good RDT can reliably detect parasitemia as low as 100-200 parasites/ml, which

is comparable to the sensitivity of regular well performed optical microscopy (Bell et al., 2006). The RDT methods are first approved by FDA approved in 2007. Now, WHO recommends RDT as the first test method in all malaria-endemic areas around the world. However, it is recommended that the results of all RDT tests be confirmed by a microscopic blood analysis and, if come positive, confirm the species and quantify the proportion of infected red blood cells (WHO 2015).

5.3 Molecular tests

The polymerase chain reaction (PCR) is another method for detecting malaria. When detecting malaria, this PCR method is more sensitive and specific than all conventional methods. PCR tests confirmed the presence of parasitic nucleic acids, and it can detect less than 1 parasite / μL (Mathison et al., 2017). PCR results are usually not fast enough to be used for diagnosing malaria in endemic areas. However, PCR is more useful to confirm the malaria parasitic species after performing the diagnosis of smear microscopy or RDT. Furthermore, PCR is useful for monitoring patients receiving antimalarial treatment (Mathison et al., 2017; Rougemont et al., 2004).

5.4 Indirect immunofluorescence assay and ELISA test

Indirect methods of malaria detections are based on an indirect IFA or an ELISA test which are used to demonstrate antibodies to malaria-causing agents. It is a sensitive way to demonstrate past exposure to malaria parasites (CDC 2018). If antibody IGM is present, that indicates present infection. On the other hand, presence of IGG antibody indicates past infection (such as 1 or 2 years previous infection). However, these methods are not used for routinely diagnosis. But these methods may be useful evaluation of treatment method and screening of blood donors prior to blood transfusion to avoid infection (kim et al., 2003).

Chapter 6

Malaria Treatment

In China, a sweet sagewort plant known as Qinghai (Latin Artemisia annua) was used to treat malaria as early as the 2nd century BC (Hsu et al., 2006). Later, Different antimalarial drugs like quinolines, antifolates, and artemisinin-combination therapies (ACTs) are used to treat malaria. All of these drugs target the intraerythrocytic stage of the parasite life cycle, when symptoms are first detectable.

Quinolines incudes basic aromatic compound like quinine, chloroquinine, mefloquine, and amodiaquine. They are the oldest class of antimalarial drugs. In the 16th century, Spanish invaders in Peru took over the cinchona malaria treatment made from the Cinchona tree's bark (Latin Cinchona succirubra). In 1820, French chemists Pierre Joseph Pelletie and Joseph Bienaimé Caventou discovered the active ingredient quinine from this plant, which had been employed in the chemoprophylaxis and treatment of malaria for many years. The mechanism of action of Quinine is not very clear but it has found that, quinine gives the similar mechanism of action like chloroquine. The mechanism of action of Chloroquine is, it prevents the polymerization of heme. Heme is converted to hemozoin by undergoing polymerization. Chloroquine binds to pure heme and associates with heme-containing Plasmodium fractions in vitro, presumably competing with free heme and preventing the conversion of heme to hemozoin (Sullivan et al., 1996). Thereby, it increases the level of the heme which is toxic to the plasmodium parasite. However, Plasmodium must convert heme to a non-toxic form because it is toxic to the parasite. Although the mechanism for this conversion is unknown (neither humans nor mosquitos generate hemozoin), it is an excellent pharmacological drug

retroactively. Quinoline-resistant strains are thought to have mutations in genes producing proteins required for drug transport into Plasmodium's food vacuole, and at least two known vacuole membrane transporters have been shown to be altered in chloroquine-resistant strains (Le Bras et al., 2003). Hemozoin inhibitors are assumed to be present in all quinolines.

Current malaria treatments rely heavily on ACTs. Since the late 1970s, artemisinins, which are derivatives of the Chinese herb Artemisia annua, have been utilized (Rogerson et al., 2010). Basically, artemisinin drugs are mostly used as prodrugs, which are initially inactive and then they are converted to active form when they reach to liver after metabolism. Artemisinin is activated by hydrolysis to the metabolite dihydroartemisinin. Antimalarial activity of artemisinin medicines is mediated by the formation of a radical via a peroxide linkage (Guo 2016). Besides, they are sesquiterpene lactones that reduce parasite load much faster than conventional antimalarial medications, and they can also kill Plasmodium gametes, lowering transmission rates. (Hyde 2002) The mechanism of action of ACT is unknown, however they are thought to prevent the conversion of heme to hemozoin, similar to quinolones (Hyde 2002). Also, Because of the high resistance to chloroquine, sulfadoxine-pyrimethamine, and amodiaquine, ACT treatments are utilized mostly. Again, Artemisinin-resistant Plasmodium strains have already emerged, as they have with other antimalarial, however ACTs improve the chances of containing these resistance variants. According to WHO (2000), to achieve a high cure rate of P. falciparum malaria and minimize the emergence of drug resistance, WHO recommends the use of artemisinin-based combination treatments (ACT).

Later, DDT was widely used as antimalarial drug in Second World War (Meshnick et al., 2001) .DDT is an organochlorine insecticide that was used to kill insects in liquid and powder form. People were sprayed with DDT during World War II (Ray 2010). Two-thirds of the world's population had been exposed to malaria from the late middle Ages until 1940, when DDT was

introduced, posing a serious health, demographic, and economic concern. (Meshnick et al., 2001) (Flannery et al., 2013), (Zhao et al., 2016)

Numerous efforts have been made over the last several decades to create effective and affordable antimalarial vaccines. Several clinical trials have been performed in recent years. Clinical trials for the development of next-generation malaria vaccines are now underway. The biggest concern is the *P. vivax* vaccine, which requires more research to develop novel vaccine candidates. (Karunamoorthi et al., 2014). Recently, The World Health Organization (WHO) recommends that children in Sub-Saharan Africa and other places with moderate to high P. falciparum malaria transmission get the RTS,S/AS01 (RTS,S) malaria vaccine. The proposal is based on the outcomes of a trial program that has served over 800,000 children in Ghana, Kenya, and Malawi since 2019. The RTS,S/AS01 malaria vaccine should be given to children starting at the age of 5 months on a four-dose schedule to reduce malaria disease and burden. (WHO 2021)

We have summarized the most common current drugs that has been using from the beginning, also summarized its mode of action, usage and advantages or disadvantages that effect the treatment of malaria in table 1.

Table 1: Overview of most common antimalarial drugs

Name of the drugs	Origin	Discovery/ Synthesis	Usefulness	Mode of action	Side effects	Advantages\ Disadvantages
Quinine	Cinchona tree, South America	1600	Resistance to chloroquine, prophylaxis and treats malaria	Inhibits DNA and RNA synthesis.	Avoided during pregnancy	Poisonous, less effective
Chloroquine	Synthesized by German scientist Hans Andersag	1934	The most potent prophylactic remedy. treats malaria	Inhibits DNA, RNA synthesis	Irritation of the skin, headaches, gastrointestinal difficulties	Resistance development against most P. falciparum strains
Primaquine	The 8-aminoquinoline derivative	1953	Infections caused by *P. vivax* and *P. ovale*, malaria prevention and treatment	Electron transport chain is interfered also parasite mitochondria is destroyed	Anorexia, nausea, anemia, headache	Relapse caused by P. vivax and P. ovale infection is prevented
Doxycycline	Pfizer Inc. New York	1960	Prophylaxis against mefloquine-resistant *P. falciparum.*	It binds to 30S ribosomal subunit and inhibits protein synthesis	Gastrointestinal disorders, nausea, vomiting, photosensitivity	Cheaper and much effective
Mefloquine	USA army and WHO	1971	Multi resistant *P. falciparum* strains, and malaria treatment	Damage to parasite membrane	CNS disorder, contraindicated in pregnancy and patients with epilepsy	Partial resistance, brain damage
Proguanil (chloroguanide)	Biguanide derivate	1953	Prophylaxis in infections with *P. falciparum.*	Inhibits DNA synthesis	If doses taken for longer time, it may cause digestive problems	It is the least toxic antimalarial drug
Atovaquone/ Proguanil	Ubiquinone analog	2000	Used For the prophylaxis and malaria treatment	cytochrome bc1 is inhibited in Plasmodium	Nausea, vomiting, diarrhea, headache, dizziness, anxiety	Widely used, lesser side effects and more expensive than mefloquine, *P. falciparum* resistance

Chapter 7

Conclusion

Malaria, which is spread by mosquitos and caused by the parasite P. falciparum, kills millions of people each year in tropical and subtropical places around the world. So, it is now crying need to develop new treatments and invest more in vaccine development research that can significantly and reliably interrupt the life cycle of this complex parasite. Small temperature increases in cold climates could significantly increase the risk of malaria transmission, allowing vectorial capacity to rise above a threshold where malaria transmission could be sustained. Further, many other factors are also important in case of malaria transmission, including socio-ecological and environmental factors such as agricultural practices, deforestation, water storage and disposal systems, population density, living conditions, health infrastructure, human behavior, immunity of human beings and the levels of drug resistance of the parasite (Reiter, 2001) . However, it is not possible to eliminate malaria parasite or malaria vector completely. But it can be controlled by following some necessary steps such as preventing insect bites, Eliminating mosquito habitat and destroying mosquito larvae.

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