1. INTRODUCTION

Malaria is a disease caused by parasite of the genus *Plasmodium* and it is transmitted via the bites of infected female mosquitoes of *Anopheles* species. *Plasmodium falciparum* and *Plasmodium vivax* are the rampant species, but the infection caused by the former is the deadliest. At present, there are around 250 million cases of malaria world-wide. The majority of cases are prevalent in the African zone, South-East Asia and Eastern Mediterranean zones (World Malaria Report, 2008). The individual most at risk of significant morbidity and transience on account of malaria are the children and the pregnant women (Ashley *et al.*, 2006; Laloo *et al.*, 2006).

The term “malaria” derived from the Italian words ‘mala’ meaning bad and aria - meaning ‘air’, was used by Dr. Francisco Torti, when people associated the disease with foul air of marshy land. It was later in 1880 that a French Army Physician, Laveran showed that malaria is caused by *Plasmodium* protozoan. Later, Ronald Ross, a British Army Surgeon in India, demonstrated that the parasite disease is transmitted by ‘anopheles’ mosquito (Parry, 2005). This discovery earned him Nobel Prize in 1902 (Enayati and Hemingway, 2010).

Human malaria is caused by *Plasmodium falciparum*, *Plasmodium vivax*, *Plasmodium ovale* and *Plasmodium malariae*, each of which differs in geographical distribution, microscopic appearance, clinical features and potential for the development of resistance to the antimalarial drugs. New empirical estimates put the number of episodes of clinical *Plasmodium falciparum* malaria in the region of half a billion per year. The mortality rate from malaria has been estimated at approximately 2.7 million per year. It is shocking to note that more than one million children die each year in Africa from malaria.
1.1 LIFE CYCLE OF MALARIAL PARASITE

All *Plasmodium* species have two hosts, vertebrate and mosquito. Malaria is caused by the parasites (sporozoites), passed into the human circulation by the bite of infected anopheles mosquitoes. The sporozoites get lodged into parenchymal cells of the liver, where they multiply and develop into tissue schizonts. This forms the primary erythrocytic or pre-erythrocytic phase of infection, and it lasts for 5 to 16 days, depending on the species of plasmodium. Thousands of merozoites are released upon rupturing of tissue schizonts. The merozoites enter circulation and invade erythrocytes to start the erythrocytic phase or blood cycle (Figure 1).

![Figure 1: Life Cycle of Malarial Parasite](image)

The secondary exo-erythrocytic phase involves a portion of these parasites infecting more liver cells. Some tissue parasites may remain dormant (latent forms or hypnozoites) in *P. vivax* and *P. ovale* infections, but not in *P. falciparum* and *P. malariae* infections. The dormant parasites may result in relapse, months or years later in the infected patient. Most parasites undergo asexual development in erythrocytes through trophozoites and finally mature to schizonts. The schizont –
INTRODUCTION

containing erythrocytes on rupturing release 6-24 merozoites. There is a feeling of chill and fever when erythrocytes burst. The liberated merozoites infect more red blood cells and start the cycle afresh; this continues until death of the host or modulation of the drug or acquire immunity. The periodicity of fever in tertian or quartan malaria is therefore based on the timing of schizogony of a generation of erythrocytic parasites.

Some of the merozoites differentiate into male and female parasites known as gametocytes. These gametocytes can undergo sporogony (sexual cycle) in the gut of female mosquito. The resulting zygote develops in the gut wall as an oocyst and ultimately gives rise the infective sporozoites which invade the salivary gland of the mosquito (Singh and Kapoor, 2012)

1.2 DRUGS USED IN THE TREATMENT OF MALARIAL PARASITE INFECTION

Antimalarial drugs are used in the treatment and prevention of malaria. However, in the chemotherapy of malaria the emergence of resistance to the available drugs is the major obstacle. The acquired drug resistance is a serious clinical problem only with *P. falciparum*, which accounts for 85 % of the cases and much of the mortality of human malaria. The major classes of drugs which have been used or are being used are discussed under the heads (Singh and Kapoor, 2012):

1. QUININE AND OTHER AMINO ALCOHOLS
2. 8-AMINOQUINOLINES
3. 9-AMINOACRIDINES
4. 4-AMINOQUINOLINES
5. BIGUANIDES AND AMINOPYRIMIDINIES
6. SULPHONAMIDES
7. ARTEMISININ AND DERIVATIVES
1.2.1 QUININE AND OTHER AMINO ALCOHOLS

Quinine (1) is the main alkaloid of cinchona bark. Preparations from the cinchona bark were used in Europe as a cure for malaria. Therefore, cinchona got official recognition in the London Pharmacopoeia in 1677. It led to the successful cultivation of the trees in Java, Indonesia and India in around 1880. The main alkaloid was isolated in 1820, structure was determined in 1908 and 1944. Several of the eminent chemists have attempted its total synthesis. However, the procedures have been too complex and not commercially viable. Quinine (1) and related alkaloids were the only antimalarial drugs until 1932.

\[
\begin{align*}
\text{OCH}_3 \\
\text{N} \\
\text{Ar} \quad \text{R} \\
\text{C} \quad \text{R}_1 \\
\text{N} \quad \text{R}_2 \\
\text{C} \quad \text{CH} \quad (\text{CH})_n \\
\text{OH}
\end{align*}
\]

From the structure activity relationship it was observed that all the four alkaloids show antimalarial activity but their C-9 epimers were found to be inactive. Any changes in the secondary alcoholic function at C-9 diminishes the activity. However, an alkyl tertiary amine linked to C-9 is necessary. This observation led to study of different amino alcohols having the following structure (2):
INTRODUCTION

At the Walter Reed Institute for Medical Research, there was established Malaria Research Programme in 1963 for developing compounds which could be effective against the strains of the drug – resistant *P. falciparum*. Two promising drugs: Mefloquine (racemate of active (3) and (4)) and Halofantrine (5) have emerged out of this work. Mefloquine is useful for the prevention of malaria in all areas except for those where parasites have resistance to multiple drugs.

![Mefloquine](3)

![Halofantrine](4)

Halofantrine (5) is a 9-phenanthenemethanol, acts as a blood schizontocide effective against all plasmodium parasites (Wesche et al., 2000).

![Halofantrine](5)

Lumefantrine (6) is an antimalarial drug and used in combination with artemether (Arinaitwe et al., 2009).

![Lumefantrine](6)
1.2.2 8 - Aminoquinolines

P. Guttmann and P. Ehrlich (1891) examined the dye methylene blue (7) for its antimalarial potential and observed that the dye had some beneficial effects on patients suffering from malaria. Several methylene blue related analogues were prepared in which one of the methyl groups was replaced by a dialkylaminoalkyl group. There were obtained some more active compounds which led to the conclusion that dialkylaminoalkyl side chain provided enhanced antimalarial activity. Therefore, it was thought to prepare dialkylaminoalkyl analogues of 6 – methoxyquinoline moiety of quinine. This research culminated in the introduction of first synthetic antimalarial drug pamaquine (plasmoquine) (8). It was active as tissue schizontocide.

Several analogues of 8-aminoquinoline were prepared and evaluated for antimalarial activity. Out of these primaquine (9) has emerged as clinically useful antimalarial drug. Primaquine kills primary and secondary exo-erythrocytic forms of the plasmodia.
It also has a marked gametocytocidal activity against all the four species of plasmodia that infect man, especially *P. falciparum*. It has little action on erythrocytic stage and as such should not be used alone in the treatment of malarial attack. Primaquine (9) is mainly used for the radical cure of *vivax* and other relapsing malaria.

### 1.2.3 9-AMINOACRIDINES

A large number of acridine derivatives containing basic side chain together with other substituents were prepared and evaluated. These investigations resulted in the discovery of quinacrine (mepacrine) (10) in 1932, the first synthetic antimalarial drug with blood schizontocidal activity. Structurally, quinacrine has the 6-methoxyquinolyl moiety of quinine (1) and the basic side chain of pamaquine (8).

![Chemical Structure of Quinacrine](image)

Numerous other related 9-aminoquinolines were designed and tested but none was found to be better than quinacrine.

### 1.2.4 4-AMINOQUINOLINES

4-Aminoquinolines are structurally related to 9-aminoacridines, which may be considered to be wonderside of two 4-aminoquinolines. These were first explored in Germany. Sontoquine (11) was selected out of these for the field studies in North Africa during World War II, where samples were captured by American soldiers. Structure of the Sontoquine (11) was determined which led to the study of several 4-aminoquinolines in USA. Among the series, Chloroquine (12) was found to be most effective antimalarial drug.
INTRODUCTION

However, chloroquine (12) had been studied in Germany in 1934 under the name Resochin but had been dropped as it was considered toxic. Chloroquine (12) is a fast – acting schizontocide. Hydroxychloroquine (13) is related to the chloroquine in which one of the N-ethyl substituent is β-hydroxylated. Its antimalarial activity is essentially equivalent to chloroquine.

The findings that certain phenolic biphenyl Mannich bases \([\text{ArCH}_2\text{N(C}_2\text{H}_5)_2]\) exhibited antimalarial activity prompted studies on compounds having related kind of side chain at position 4- of quinoline. These studies resulted in the emergence of many useful agents, especially amodiaquine (14). The other related analogue is cycloquine (15) which was prepared in Russia.
INTRODUCTION

Hydroxypiperaquine (16) from China was claimed to be active against chloroquine-resistant strains (Singh and Kapoor, 2012).

1.2.5 BIGUANIDES AND AMINOPYRIMIDINES

In the forties, work in Britain led to the discovery of Proguanil (17), a research team in America at the Wellcome Research Laboratories studied 2,4-diaminopyrimidines as anti-folate agents. 2,4-Diamino-5-(p-chlorophenoxy)pyrimidine (18) was found to antagonize folic acid in Lacto bacillus cases in vitro. There is striking resemblance between Proguanil (17) and 2,4-Diamino-5-(p-chlorophenoxy)pyrimidine (18) written next in a way so that analogy is evident, which also has anti-folate properties. Thus, diaminopyrimidines were screened as antimalarials. Compound was found to be active in animal tests.
INTRODUCTION

The pursuance of this line of thought, first the isosteric replacement of ether oxygen by CH₂ and later the deletion of any bridging atom between the two rings, gave the anti-folate pyrimethamine (19), which is well established as an antimalarial.

Later, while carrying out work on 2,4-diamino-5-benzylpyrimidine type of antifolates, trimethoprim (20) was created as antibacterial but subsequently found to have antimalarial properties (Singh and Kapoor, 2012).

1.2.6 SULPHONAMIDES

Sulphonamides have inhibitory effect on the erythrocytic phase of \textit{P. falciparum}. These drugs have been employed in combination of pyrimethamine for fast action. Sulfadoxine (21) and sulfamethopyrazine (22) are ultra-long acting sulfonamides, show synergistic action with pyrimethamine and found to be active
against chloroquine resistant *P. falciparum*. Resistance to sulfa-pyrimethamine has been observed to a limited extent in India and only few cases have been confirmed which are restricted to North Eastern states.

![Chemical structures](image_url)

However, the resistance to this combination has been observed in South East Asia, South America and Southern Africa. Therefore, quinine has also been prescribed in such cases. Further, this combination has not been found to be active against *vivax* malaria (Tripathi K. D., 2003; Singh and Kapoor, 2012).

### 1.2.7 ARTIMISININ AND DERIVATIVES

Artemisinin (23), a natural compound from *Artemisia annua* (sweet wormwood) plant, is highly effective against drug-resistant malaria and employed in Chinese traditional medicine. Artemisinin is a sesquiterpene lactone containing an unusual peroxide bridge. This peroxide is believed to be responsible for the drug’s mechanism of action (Woo et al., 1998).

![Chemical structure of artemisinin](image_url)

Semi-synthetic artemisinin derivatives (e.g. artesunate (24), artemether (25)) are easier to use than the parent compound and are converted rapidly once in the body to the active compound dihydroartemisinin. Artesunate is a semi-synthetic derivative of artemisinin that is water-soluble and may be given by injection (Noston et al., 2000).
Dihydroartemisinin (26) is the active metabolite of all artemisinin compounds and is also used as a drug in itself. Arteether (27) is an ether derivative, used in combination therapy for cases of uncomplicated resistant *P. falciparum* (Arinaitwe *et al.*, 2009).

The mechanism of antimalarial action of artemisinin analogues is mediated by free radicals (Knmgkrai and Yuthavong, 1987; Levander *et al.*, 1989; Meshnick *et al.*, 1989). This category of antimalarial drugs contain an endoperoxide bridge necessary for antimalarial action (Brossi *et al.*, 1988) which appears to react with intraparasitic heme iron (Meshnick *et al.*, 1991). The interaction between artemisinin analogues and heme is responsible for the selectivity of the drug (Goldberg *et al.*, 1990).

Use of the artemisinin by itself as a monotherapy is explicitly discouraged by the World Health Organization, as there have been signs that malarial parasites are developing resistance to the drug. Therapies that combine artemisinin with some other antimalarial drug are the preferred treatment for malaria and are well tolerated in patients (Parry, 2005).