Introduction
MALARIA: GLOBAL SCENARIO AND LIFE CYCLE

Malaria is the accepted name denoting the disease or condition of infection caused by parasites belonging to genus *Plasmodium*. Although the name malaria, which is derived from the Italian word for bad air (mal=bad + aria=air), is not directly related to the cause of infection, it is the term commonly used, since the scientifically more appropriate term-Plasmodiose-has never come into wide use. Malaria is thought to have originated in Africa and affected prehistoric man. It is caused by infection with a pathogenic protozoan parasite of blood, the *Plasmodium*. The *Plasmodium* parasites are highly specific, with female *Anopheles* mosquitoes as the vectors (WHO, 1987). Human malaria is caused by four species of the parasite i.e. *P. falciparum*, *P. vivax*, *P. ovale* and *P. malariae*. The four species causing human malaria differ morphologically, immunologically, in geographical distribution, relapse pattern and in drug response. *P. falciparum* and *P. vivax* are the two species that contribute heavily to the malaria burden. *P. falciparum* causes the most serious disease. The most common malaria parasite is *P. vivax*; infections are rarely fatal. Least common is *P. ovale*, which is restricted to West Africa and also produces a mild illness. *P. malariae* is found in isolated places scattered across the globe, and while it causes severe fever, it is usually not life threatening. Species of the *Plasmodium* parasite are also found in primates, rodents, bats and other mammals, birds and reptiles. *P. knowlesi*, *P. cynomolgy* and *P. simium* are some species infecting monkeys; *P. yoelii*, *P. berghei* cause rodent malaria, and *P. gallinaceum*, *P. lophurae* cause avian malaria.

Malaria has long been, and still is, one of the most harmful diseases of man. It has caused widespread destruction of mankind. The global burden of human malaria is enormous, amounting to many new infections and millions of deaths annually. This is the situation more than 100 years after two key discoveries; one by Charles Laveran in 1880, that the infection is caused by a blood-dwelling apicomplexan parasite belonging to the genus *Plasmodium* and the other by Ronald Ross in 1897, that the parasites are transmitted by blood-sucking female anopheline mosquitoes (Kumar, 2002). In spite of tremendous progress in science in the present century, millions of people still die of malaria every year, especially in tropical and subtropical regions.
Malaria ranks third among the major infectious diseases in causing deaths—after pneumococcal acute respiratory infections and tuberculosis. It is expected that by the turn of the century, malaria would be the number one infectious killer disease in the world. Worldwide, about 1.5 million to 3 million people die of malaria every year (85% of these occur in African children), accounting for about 4-5% of all fatalities in the world. Between 400 million and 900 million cases of acute malaria occur annually in African children alone. WHO forecasts a 16% growth in malaria cases annually. It accounts for 2.6% of the total disease burden of the world and is responsible for the loss of more than 35 million disability-adjusted life-years each year. Previously extremely widespread, the malaria is now mainly confined to Africa, Asia and Latin America. It is a disease that can be treated in just 48 hours, yet it can cause fatal complications if the diagnosis and treatment are delayed. The problems of controlling malaria in these countries are aggravated by inadequate health structures and poor socio-economic conditions. The emergence of parasite resistance to our limited armamentarium of drugs contrives a formidable therapeutic challenge and necessitates the development of new drugs for the treatment of malaria.

LIFE CYCLE OF PLASMODIUM PARASITES AND PATHOGENESIS

The Plasmodium genus of protozoal parasites undergoes a complex life cycle, alternating between vertebrate host and arthropod vector (digenetic). It comprises several stages and requires two hosts for completion—a primary, definitive or principal host, where it undergoes asexual cycle and schizogony and a secondary, intermediate or vector host where it undergoes a sexual cycle followed by an asexual multiplication called sporogony. The mosquito is always the vector, and is always an anopheline mosquito. Only female mosquitoes are involved, as the males do not feed on blood. The basic life cycle of the parasite is shown in fig 1.
The biology of infection mechanism and life cycle of different *Plasmodium* species differ to some extent and is reflected in the pattern of the disease syndrome; *P. falciparum* and *P. vivax* cause severe anemia but *P. falciparum* additionally also causes cerebral infection, hypoglycemia, metabolic acidosis and respiratory distress (Miller *et al.*, 2002).

![Diagram of the life cycle of Plasmodium in Man and the mosquito](image)

**Fig 1: The life cycle of Plasmodium in Man and the mosquito**

The infective forms of the parasite - the sporozoites, are injected from the mosquito salivary gland into the human as the mosquito must inject anticoagulant saliva to ensure an even flowing meal. Once in the human bloodstream, these sporozoites reach the liver in 30–40 min and start developing in the parenchymal cells
of the liver. Each sporozoite multiplies into 20-30,000 merozoites, ruptures the liver cell and is liberated into the blood stream where it infects red blood cells. The liver and sporozoite stage together are termed *Pre-erythrocytic schizogony*. Since only a few hepatocytes are affected, this phase does not produce any symptoms or signs. The merozoites in the blood infect red blood cells and develop within these cells through stages. Each merozoite can have two fates; it can either divide into 8-32 fresh merozoites, which rupture the red cell and infect fresh red cells. This cycle occurs every 48-72 hours and it is called *erythrocytic schizogony*. Alternatively, merozoites can produce micro and macrogametocytes, which have no further activity within the human host. All the clinical manifestations of malaria are due to the erythrocytic phase that results in the release of pro-inflammatory products of red cell membrane and of the parasites. The clinical manifestations like chills and fever coincide with this 48-72 hours cycle and occur every alternate day or once in 72 hours. Some of the tissue forms in *P. vivax* and *P. ovale* go into hibernation (to tide over the adverse conditions in the sub tropical and temperate regions) and get re-activated once in 2-6 months to cause relapse of symptoms. Another mosquito arriving to feed on the blood may suck up the sexual forms-micro and macrogametocytes (male and female) into its gut, where exflagellation of microgametocytes occurs, and the macrogametocytes are fertilized (WHO, 1987). The resulting ookinete penetrates the wall of a cell in the midgut, where it develops into an oocyst. Sporogeny within the oocyst produces many sporozoites and, when the oocyst ruptures, the sporozoites migrate to the salivary gland, for injection into another host.

With the first attack of *P. falciparum*, fever is usually irregular rather than occurring with a regular, repeating pattern as seen with a tertian fever in subsequent attacks, and there are usually no relapses unlike with *P. ovale* and *P. vivax* where hypnozoites are formed (Peters *et al.*, 1995). The temperature may rise above 41°C, and the fever is produced as the schizonts mature, at 48 hr intervals usually for *P. falciparum*. Malaria is especially dangerous to pregnant women and small children. Symptoms include headache, fatigue, fever and aches, diarrhoea and abdominal pain (WHO, 1991). Severe and complicated malaria is usually caused by delay in treating an uncomplicated attack of *P. falciparum*; a patient with severe and complicated malaria will often present with impaired consciousness, weakness, and jaundice.
Other complications are cerebral malaria (unrousable coma), generalized convulsions, anaemia, renal failure, hypoglycaemia, electrolyte and acid-base disturbances, pulmonary edema, circulatory collapse, hyperparasitaemia, and malarial haemoglobulinurea. These features may occur singly or in combinations.

Control of malaria involves various measures directed at reducing the load of infection in the community and reducing the transmission by controlling the mosquitoes: early diagnosis and prompt treatment, personal protection, repellants, bed nets and chemoprophylaxis.

Treatment of malaria involves administration of a blood schizonticidal drug for alleviation of symptoms and tissue schizonticidal and gametocytocidal drugs for preventing relapse and transmission respectively. Chloroquine is the most effective, safe and cheap blood schizonticidal drug; however, resistant strains have now evolved. Chloroquine is also gametocytocidal against \textit{P. vivax} malaria and therefore, presumptive treatment with chloroquine helps in preventing the spread of \textit{vivax} infection. In cases of \textit{P. falciparum} infection with proven chloroquine resistance, second line drugs like quinine, pyrimethamine + sulfadoxine or tetracyclines can be used. In cases of multi-drug resistant \textit{P. falciparum} infection, either mefloquine, halofantrine or artemisinin derivatives can be used.

The control of malaria is a formidable task. Due to various reasons, malaria control programmes received setbacks all over the world and today it has come back with a vengeance, and with new features not witnessed during the pre-eradication days: vector resistance to insecticide (s), resistance in \textit{P. falciparum} to chloroquine and other antimalarial drugs and human resistance to chemical control of vectors. Malaria control has become a complex enterprise, and its management requires decentralization and new approaches. At this stage, target specific drugs should be designed and synthesized by using combined knowledge of classical metabolic biochemistry, combinatorial and computational chemistry or molecular modeling.
FILARIASIS: GLOBAL SCENARIO AND LIFE CYCLE

Among the helminthic infections, filariasis or elephantiasis is the most debilitating and disfiguring scourge among all diseases. Filariasis is one of the major public health and socio-economic problem tropical and subtropical world are facing; it is a mosquito-borne parasitic disease caused by three species of tissue dwelling filaroid nematodes. *Wuchereria bancrofti* is responsible for 90% of cases and is found throughout the tropics and in some sub-tropical areas worldwide. *Brugia malayi* is confined to southeast and eastern Asia. *Brugia timori* is found only in Timor and its adjacent islands. Filariasis has a long history, which reaches back into antiquity (Routh and Bhowmik, 1993). Despite this, filariasis has been a disease, which up to recent times, has been poorly understood and largely ignored by health authorities that were struggling to control what were perceived to be more important vector-borne diseases such as malaria and dengue fever. In the last two decades or so there has been a flurry of filariasis research, which has provided new insights into global burden of filariasis, the pathogenesis of filarial disease, diagnosis, and control. In addition to the medical problems, there are severe social and psychological consequences associated with this disease, especially in those who suffer from elephantiasis or hydrocoele. Filarial diseases are still far from being extinct, thus pose a challenge for multiple areas of research.

Filariasis has afflicted people in the tropical areas of the world for thousands of years but even up to comparatively recent times it has been poorly understood and its importance under-recognized. Filarial nematodes infect more than 150 million humans worldwide and threaten one billion individuals in Africa, India, Southeast Asia and America. While filariasis is endemic in 80 countries, an estimated 70% of infected cases are concentrated in India, Nigeria, Bangladesh and Indonesia. Among the 38 least developed countries, 32 are endemic for filariasis. In India, an estimated population of 22 million is known to harbor circulating microfilariae and 16 million people suffer from filarial manifestations like elephantiasis of limbs, genitals and hydrocoele.

The thread-like, parasitic filarial worms *W. bancrofti* and *B. malayi* that cause filariasis live almost exclusively in humans. These worms lodge in the lymphatic
system, the network of nodes and vessels that maintain the delicate fluid balance between the tissues and blood and are an essential component for the body's immune defence system. Each adult worm lives for six years in the lymphatic system, and female worms release millions of microfilariae (immature worms) that circulate in the blood. In India, *Mansonia* spp. of mosquitoes propagates *B. malayi* whereas *Culex pipens* is responsible for *W. bancrofti*. The World Health Organization (WHO) has identified lymphatic filariasis as the second leading cause of permanent and long-term disability worldwide.

**LIFE CYCLE OF FILARIAL PARASITES AND PATHOGENESIS**

The pathological symptoms of filarial infection can be broken down into the following categories: lymphatic filariasis, subcutaneous filariasis, and serous cavity filariasis. There are three distinct phases in the life cycle of filarial parasites: microfilariae, infective larvae and adult worms (Fig. 2). The adult worms live in lymphatic, connective tissue/vessels or cavities of the host and release microfilariae (sheathed/unsheathed) into peripheral blood of the host. Vertebrates are the definitive host whereas female mosquito is the intermediate host. The transmission is affected by the uptake of the first stage larvae (microfilariae, mf) into hematophagous arthropods as they feed on the infected host. The mf then migrate out of the digestive tract into the hemocoel, and in suitable locations (frequently the thoracic muscles) develop without multiplication into second and later on into more advanced third stage larvae. The mature larvae migrate into the hemocoelic cavity in the labium and escape into the vertebrate host's skin while circulating in peripheral blood, some mf are taken up by insect vector (mosquito) in which they undergo further development to the infective larval stage within 15 days. Microfilariae have a life span of 14-70 days and exhibit nocturnal/diurnal periodicity. When the blood sucking arthropod takes its next meal, the infective larvae are transmitted to the recipient host through skin and thus the cycle is completed. This process takes place over a period of several months during which the parasite moults twice. There are no free-living stages and both the microfilariae and the adults may persist for a long time, many years in case of adult worms.
The disease is due to host's immune response to the worms, particularly dying worms. Pattern and severity of the disease varies with the site and stage of species. The adult worms survive for about 10-15 years. The infections are chronic and worst in individuals constantly exposed to reinfection. Filarial infections cause the highest eosinophilia of all helminthic infections, and are normally caused by the morphology of the microfilariae.

**External manifestations:** The adult worms damage the lymphatic system, causing fluid to collect and cause swelling in the arms, legs, breasts and genitals; this condition is known as lymphoedema. Acute episodes of local inflammation involving skin, lymph nodes and lymphatic vessels often precede or accompany lymphoedema. Some of these are caused by the body's immune response to the parasite, but most are the result of bacterial infections of skin where normal defences have been partially lost due to underlying lymphatic damage. Such infections cause a grotesque hardening and thickening of the skin, known as elephantiasis. Millions suffer from debilitating acute attacks of filarial fevers and lymphadenitis. In endemic communities, 10-50% of men suffer from evident clinical disease--lymphedema, genital damage, especially hydrocoele (fluid-filled balloon-like enlargement of the sacs around the testes) and elephantiasis of the penis and scrotum and recurrent infections associated with damaged lymphatics, lung disease, chyluria and renal disease. Elephantiasis of the entire leg, the entire arm, the vulva, or the breast can affect up to 10% of men and women in these communities.

Many people with lymphatic filariasis never develop clinical signs of the infections. However, studies show that such people actually have hidden internal (preclinical) damage to their lymphatic and renal systems.
There are two principal goals for programmes to eliminate filariasis:

1. To interrupt transmission of infection: The essential strategy is to treat the entire 'at risk' population for a period long enough to ensure that levels of microfilariae in the blood remain below those necessary to sustain transmission. For the yearly, single-dose, 2-drug regimens being advocated (Albendazole: 400 mg + DEC: 6 mg/kg, or albendazole: 400 mg plus ivermectin: 200 mg/kg); this period has been estimated to be 4-6 years, corresponding to the reproductive lifespan of the parasite. For the treatment regimen based on the use of DEC-fortified salt, the period has been found empirically to be 6-12 months of daily-fortified salt intake.

Fig 2: The life cycle of Filarial worms in Man and Mosquito

2. To alleviate and prevent both the suffering and disability caused by the disease: the principal strategy focuses on decreasing secondary bacterial and fungal infection of limbs or genitals whose lymphatic function has already been compromised by filarial infection, since it is this secondary infection that has recently been identified as the primary pathogenetic determinant of worsening lymphoedema and elephantiasis.
Operationally, a regimen of meticulous local hygiene to affected areas and the creation of hope and understanding among the patients and their communities are the principal strategic approaches.

At present no effective and safe chemotherapy is available against the adult human filarial parasites, which raises an urgent need for the development of macrofilaricidal drugs. The successful treatment of filariasis, a disease of many tropical and subtropical areas, is not possible because of the non-availability of macrofilaricidal drugs (National workshop NICD, 1991; Ottesen and Ramachandran, 1995; Cao et al., 1997). The age-old drug diethylcarbamazine (DEC), introduced in 1947, continues to be the mainstay of clinical practice despite its well-known deficiencies (Sharma, 1990; Simonsen et al., 1997). Suramin, levamisole and mebendazole, the DEC analogue centperazine, and ivermectin are also suggested for filarial treatment, but all of them show severe toxic side effects. Ivermectin, a semisynthetic macrocyclic lactone antibiotic, may take an impact as microfilaricide for onchocerciasis, but did not irreversibly damage the adult filarial worms (Bennett et al., 1988). Although organic arsenical compounds have long been known as good macrofilaricides (Ginger, 1986), their potential toxicity to the host has prevented their development as useful antifilarial drugs. Besides these antifilarials, a number of phenoxy-cyclohexane derivatives (Loiseau and Depreux, 1993), 2,4,6-substituted triazines (Chauhan and Chatterjee, 1994), 5-amino and 5,8-diaminoisoquinolines (Srivastava et al., 1996), aplysinospin derivatives (Singh et al., 1997a) and 1,10-dicyano-2-substituted ethylenes (Tewari et al., 1997) were identified as potential filaricides but most of the compounds exhibited very poor adulticidal response. Benzimidazole group of anthelmintics exhibits high order of activity against intestinal helminths but has not found application for the treatment of tissue dwelling helminths (Sharma, 1994; Townsend and Wise, 1990). Therefore, the need arose to identify structural prototypes associated with macrofilaricidal activity. Though annual mass treatment with DEC either alone or in combination with ivermectin and albendazole has been proven to be very effective in destroying microfilariae, but the ultimate goal of finding a safe, effective and affordable macrofilaricide still eludes us.