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New answers through chemotherapy?

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Introduction

In 1898 Manson[22] wrote 'Many drugs have been employed in the treatment of malarial disease, and many drugs have some influence on it; all sink into insignificance in comparison with quinine.' Today, 85 years and over a quarter of a million drugs later one could make almost the same statement! What roles should drugs perform in the containment of malaria, to what degree has the assortment of drugs in current use filled these roles, what are their shortcomings and what is being done to improve the situation? These are the questions that will be addressed in this paper. Before approaching these questions, however, let it be emphasized that a drug never has nor ever will serve other than as one of the major weapons in the fight to eliminate malaria as a cause of human suffering and death at the community level, albeit it that specific antimalarial drugs will continue to be essential to relieve illness and save life in a high proportion of those individuals who harbor the infection.

What should antimalarial drugs do?

There are 2 quite distinct ways of considering this topic. I believe that much of the difficulty that we encounter with antimalarial drugs today is due to our failure in the past to distinguish between the role of drugs in the individual and their role in the community. This lack of discernment (which applies, incidentally, to many drugs other than antimalarials) can be traced right through the history of malaria chemotherapy in this century, for example to Koch's recommendation of quinine for the control of malaria in village communities in German New Guinea[11].

Drugs in the individual

Differences in the life cycles of the 4 species of *Plasmodium* that infect man (reviewed by Verhave and Meis, this issue), and certainly in the different basic sensitivities of the different parasites to the available drugs[26] dictate to some degree which compounds can be deployed. However, the *functions* that antimalarias are required to carry out are as follows.

Causal prophylaxis. Strictly speaking this should imply the total prevention of the infective stages, the sporozoites, from developing into pre-erythrocytic schizonts or, in *P. vivax* and *P. ovale*, hypnozoites. In practice the term is taken to mean the blockage of development of these intrahepatic stages. Primaquine which fills all three roles is, unfortunately, too toxic to be deployed as a causal prophylactic. Proguanil and pyrimethamine effectively block pre-erythrocytic schizogony, certainly of *P. falciparum* and *P. malariae* and probably of *P. vivax* and *P. ovale*. It is questionable whether they influence the establishment and survival of hypnozoites of the latter two species when used in the normal manner. On the other hand, the use of a repository preparation of the active metabolite of proguanil (cycloguanil as its embonate salt) completely protected 12 of 24 volunteers against sporozoite-induced vivax malaria, which suggests that the hypnozoites may also have been destroyed in these individuals[9].

Suppressive treatment. This, as defined in a recent WHO monograph[5], is 'treatment aimed at preventing or eliminating clinical symptoms and/or parasitemia by the early destruction of erythrocytic parasites.' Taken to its logical conclusion, continued suppressive treatment can lead to *suppressive cure*, at least in *P. falciparum* and *P. malariae* if medication is continued after leaving an endemic area for sufficient time to ensure that any developing pre-erythrocytic schizonts have matured, there being no hypnozoites in these infections. A period of 4–6 weeks usually suffices for this purpose. 4-Aminoquinolines (such as chloroquine), quinine and mefloquine are excellent drugs for this purpose, proguanil and pyrimethamine less so, although pyrimethamine together with an appropriate sulphonamide is very effective.

Radical cure. This term has caused some confusion since, by definition, it implies the 'complete elimination of malaria parasites from the body by means of continuous suppressive treatment'[5]. In the cases of *P. falciparum* and *P. malariae, radical cure* can be equated with *suppressive cure* (leaving aside, for the moment, any question of drug sensitivity or resistance). With *P. vivax* and *P. ovale* this is not so unless *suppressive treatment* is continued for several years after the individual leaves the endemic area. In these infections radical cure demands the administration of a tissue schizontocide (hypnozoitocide is an appropriate, if rather clumsy term) to eliminate the hypnozoites that are responsible for true relapses of these parasites in the blood, sometimes years after the initial infection. At the present time the only compound used for this purpose is the 8-aminoquinoline, primaquine.

Were it not for the question of drug resistance there would be little problem today in protecting or treating the individual. Resistance of *P. falciparum* to antimalarials has been considered in some detail (see Doberstyn, this issue) and will be discussed again below. The erythrocytic stages of *P. vivax* are commonly resistant to proguanil and pyrimethamine, although so far no authentic example of chloroquine-resistant *P. vivax* has been reported. *P. malariae* too is known to become resistant to proguanil and pyrimethamine[26] but not to

chloroquine. The rare species, *P. ovale*, appears still to respond to all these drugs. Strains of *P. vivax* in which high doses of primaquine are required to ensure radical cure (e.g. those such as the Chesson strain in Papua New Guinea) clearly have hypnozoites that are less sensitive to primaquine than strains from other areas. This observation, in turn, provides interesting food for thought since a) we do not yet know whether it is primaquine or one of its metabolites that exerts an antimalarial action and b) it is not known whether the hypnozoitocidal action of primaquine is directed purely against the parasites or the intimate metabolic association of host cell and parasite[4].

Drugs in the community

A community is more than the sum of its parts, and this truism must be applied with particular emphasis to the community of malaria parasites within a community of human hosts. A simple numerical example will explain why.

Suppose in a community of 1000 people, 100 are harboring erythrocytic stages of *P. falciparum*, by no means an unlikely figure in a highly endemic area. If the average level of parasitemia in each infected person is 1 parasite per million red blood cells (a low figure in many areas), and each individual has an average of 8×10^{12} red cells, each will harbor 8×10^6 parasites. The number of parasites in the community will thus be some 8×10^9.

Let us now consider the rates at which mutations can be expected to occur in malaria parasites when exposed to drugs. While no precise figures are available, Bishop[3] working with clones of *P. gallinaceum* estimated that mutants resistant to metachloridine occurred at a rate between 1 in 5×10^7 and 1 in 10^9. More recent studies with rodent malaria parasites by Beale and his associates[2] and human and rodent parasites[46] based both on isoenzyme markers and responses to various drugs indicate that the blood stages of *Plasmodium* are haploid, and that mutations such as those underlying drug resistance can readily be perpetuated either as Mendelian dominant or recessive characters by hybridization during sexual reproduction in the anopheline vector. While none of these authors was prepared to commit himself to a figure, it seems likely that spontaneous single gene mutation in *Plasmodium* occur at a frequency somewhere between 1 in 10^6 and 1 in 10^{10} organisms.

If the 1000 people are given regular doses of an antimalarial drug it is obvious that there is every possibility that the changes of there being in the community a number of parasites with spontaneous resistance to that drug is remarkably high. Taking the minimum level above one might suggest 8×10^{-1} and, at the highest level, 8×10^3.

So far this argument has been a hypothetical one. When we look at the reality of practical experience when antimalarial drugs have been applied on a community basis, it would appear that our calculations greatly underestimate the risk.

Some of the most valuable longitudinal studies on the rate of development of resistance to antimalarials in the community remain those of Clyde and Shute[8] and Avery Jones[1] with pyrimethamine in East Africa. These and other data[26,28] indicate the rapidity with which the use of such antifols as proguanil or pyrimethamine, and even of the potentiating combination of pyrimethamine with sulfadoxine, on a community basis, has led to the emergence of parasites resistant to these drugs. More recent experience with chloroquine[26,28] suggests that resistance to this compound, once established in a community, extends just as rapidly as does resistance to antifols, albeit limited to *P. falciparum*. Unlike antifol resistance, however, chloroquine resistance appears to be a highly stable character[25], dominant over sensitive parasites so that the population of resistant organisms appears even to overgrow sensitive ones in the absence of drug selection pressure.

So far we have looked at the hazard of selecting drug resistance by applying antimalarial drugs in a community. What in fact would be the roles of a drug and the characteristics required for it to be of value for community, as distinct from individual administration? The guidelines for the use of antimalarials in malaria eradication or control programmes that have been laid down by expert committees of WHO have tended to become more conservative with time as experience has taught both the potential and the limitations of the few drugs that are available. In a recent monograph published by WHO, Bruce-Chwatt et al.[5] suggested that 'collective drug protection' may have the following, limited uses:

a) For the control of epidemics,
b) As a supplementary measure in control and eradication programmes for
– presumptive treatment, 'to relieve symptoms and prevent transmission until the diagnosis is confirmed and radical treatment can begin.' A blood schizontocide is given, often as a single dose, together with a gametocytocide.
– mass drug administration in localized areas under specified conditions. The difficulties attending this use are stressed. This, in fact, is by far the most controversial use for antimalarials and there is no question that the whole policy of administering antimalarials 'en masse' needs careful re-assessment.
– for 'radical treatment', by which radical cure of vivax malaria is implied, using 5–15-day courses of primaquine as a hypnozoitocide.
c) To prevent malaria due to blood transfusion.

These limited indications are a far cry from the extensive use of drugs which formed an integral part of the chloroquine-DDT eradication era of the 1950's and 1960's. Yet, controversial as it may be, what choice do, for example, the health authorities of a country such as India have in the face of several million new cases of *P. vivax* every year, but to employ chloroquine for presumptive treatment and primaquine for radical treatment on an enormous scale, in the fact of continuing malaria transmission at a high rate, and the logistic and financial difficulties facing other methods of malaria control in that huge country?

To be ideal for administration to large numbers of people in the indications outlined by WHO, a blood schizontocide or a hypnozoitocide should be

– applicable in a single, well tolerated dose for therapy
– preferably administerable by mouth

- safe in both sexes and all age groups, with a wide safety margin in case of misuse
- cheap
- capable of being administered in widely spaced doses for prophylaxis
- as little likely as possible to select resistant mutant parasites
- a true causal prophylactic

To what degree do our currently available antimalarials satisfy these criteria? Quite apart from the serious problem of drug resistance, particularly in *P.falciparum,* not a single compound fulfils all the criteria (table).

While it must be stressed that the place of antimalarials as prophylactics for mass drug administration is restricted to rather limited circumstances, in those indications it would undoubtedly be invaluable to have a compound or drug combination that, while fulfilling the desiderata set out above, in addition could be administered as a truly long-acting formulation, i.e. one that remains effective for at least 3 months following a single dose. The logistic advantages of such a preparation are obvious. The hazards are less so, however. Overriding many of the limitations that already exist in our current drugs today, moreover, is that of the rapidly increasing appearance of multiple drug resistance in *P.falciparum* on a global scale (see chapter by Doberstyn, this issue).

Current progress in antimalarial drug research

It has required 2 major wars, 2 decades of increasing drug resistance, and a world financial recession (limiting the ability of Third World countries to cope with the costs of controlling malaria) to persuade those concerned with new drug development that effort and money should be invested in a search for new and better antimalarials. Two major programmes have provided nearly all the stimulus for the trials and achievements of recent years. The antimalarial development programme of the US Army Research and Development Command, based on the Walter Reed Army Institute of Research (WRAIR) has carried out since its inception in 1964 an unprecedented volume of research involving not only the mass primary screening of over 300,000 candidate compounds, but also basic studies on the biology and immunology of malaria. From this programme have emerged mefloquine and a handful of other promising candidates, several of which are currently in preclinical or clinical trial. The UNDP/World Bank/WHO Special Programme for Research and Training in Tropical Diseases, was first launched in 1976. The role of this 'TDR' programme is to coordinate research on malaria (and certain other parasitic diseases of the Third World), and to provide the seed money that will stimulate research workers from a wide range of fields to invest effort on resolving the so-far recalcitrant problems posed by these diseases, bringing to bear the full potential of modern biomedical science. In addition, a select few commercial companies, several national institutes (notably in the People's Republic of China) and rare academic centers have elected to join the search for new and superior antimalarials.

Progress towards this target has been made in several directions.

Experimental models

New experimental models have been reviewed in detail elsewhere[27,28] and are therefore only listed here. They are as follows:

in vitro – the use of *P.falciparum* in continuous culture. Since the seminal work of Trager and Jensen[44] and Haynes et al.[17] that opened the way to culture of these parasites, several techniques have been described by which the blood schizontocidal action of compounds can be detected[12].

– the exo-erythrocytic stages of *P.berghei* and *P. vivax* have now been grown in tissue culture by several workers including Hollingdale et al.[18] and Mazier et al.[23]. Such cultures are now being developed as screens for causal prophylactic drugs. The potential value of these systems to detect hypnozoitocidal action is at present unknown.

in vivo – rodent models have proved of immense value for the detection of blood schizontocidal activity[27] and causal prophylactic action[29]. Both the South American owl monkey *(Aotus trivirgatus)*[36] and, more recently, the squirrel monkey *(Saimiri sciureus)*[35] have proved invaluable for advanced testing of drugs against both *P.falciparum* and *P.vivax,* but *P.cynomologi* in the rhesus monkey[38] remains the only valid model for the evaluation of potential hypnozoitocides. The cost and availability of all 3 monkeys are serious impediments to the advanced, in vivo evaluation of new antimalarial drugs prior to clinical trial.

In efforts to detect long-acting blood schizontocides, testing systems based on rodent malarias have been described recently[20,39].

Uses and limitations of antimalarials for 'collective drug protection'

Drug	Causal prophylactic		Blood schizontocide					Hypnozoitocide
	Active	Spaced doses	Single dose	Oral	Safe	Spaced	Cheap	Active
Proguanil	+	–	–	+	+	–	±	? –
Pyrimethamine	+	±	–	+	+	±	±	? –
Fansidar	+	+	+	+	±	+	–	? –
Chloroquine	–	–	+	+	+	–	+	–
Quinine	–	–	–	+	±	–	–	–
Mefloquine	–	–	+	+	+	+	–	–
Artemisinine	–	–	–	–	?[b]	–	?[b]	–
Primaquine	(+)[a]	–	–	–	–	–	–	+

[a] Not used for this purpose; [b] still under evaluation.

New blood schizontocides

4-Quinoline- and 9-phenanthrenemethanols

Of these series the most advanced compound is mefloquine, a 4-quinolinemethanol (fig. 1, **I**), developed originally by the WRAIR programme and recently under the auspices of the TDR programme. This compound which has an unusually long half-life in man, is in an advanced state of clinical trial particularly in areas of Asia[16] and South America[40] where multiple drug-resistant *P. falciparum* is a major problem, and in parts of Africa[13] where the problem is rapidly becoming established. As summarized in the table, mefloquine, as well as related compounds such as halofantrene (fig. 1, **II**) are solely blood schizontocides. Mefloquine, however, has a long half-life that increases its value for community use. Its main advantage is that, so far, it has proved almost 100% effective in curing infections of *P. falciparum* that are resistant to other therapy. Mefloquine cannot be administered parenterally and may therefore have to be supplemented by quinine (or quinidine[47]) for the emergency treatment of individuals suffering from cerebral or other severe forms of falciparum malaria. The other potential disadvantage of mefloquine and related compounds is that they are, in effect, analogues of quinine against which resistant strains of *P. falciparum* are now clearly emerging especially in Thailand[7]. While quinine is not used on a large scale at present, the release of mefloquine on its own for 'collective drug protection' is, for the reasons expounded above, fraught with the danger that strains of *P. falciparum* resistant to this invaluable drug will rapidly emerge. The potential for this has already been clearly demonstrated in experimental models[24, 30].

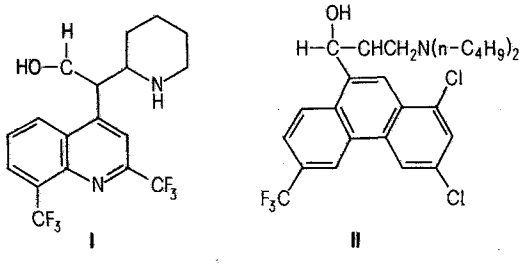

Figure 1. **I**, Mefloquine; **II**, halofantrene.

Sesquiterpene lactones

The next most promising series of blood schizontocides to be reported recently are sesquiterpene lactones based on the compound artemisinine (qinghaosu), a natural constituent of the plant *Artemesia annua* Linn. (Compositae). This unique compound (fig. 2, **I**) has been shown to have a rapid blood schizontocidal action against rodent, avian and human malarias, but no activity against hepatic stages[33]. As artemisinine itself is very poorly soluble it has had to be administered by injection but even the suspensions used to date have been shown to exert a very rapid parasiticidal action which has been of particular value in patients with cerebral malaria. This activity is maintained against parasites that are resistant to chloroquine. However, Chinese workers have noted that up to 10% of all patients treated for either falciparum or vivax infections recrudesce after the initial rapid clearance of parasitemia. The reasons for this are not yet clear, but we have found (unpublished observations) that resistance can readily be induced to artemisinine in *P. berghei* in vivo. As far as is kown artemisinine has a wide margin of safety but further toxicological evaluation of the compound is currently being carried out in China.

In view of the poor solubility of artemisinine, semi-synthetic derivatives of this lactone are being evaluated. Two of these, artesunate (fig. 2, **II**) and artemether (fig. 2, **III**) are more soluble, active and correspondingly more toxic[14]. Artesunate in particular looks especially promising for the i.v. therapy of cerebral malaria and is currently being developed for further clinical trials in this condition[14]. Meanwhile the mode of action of this series is being studied by Li et al.[21], Gu et al. [15], and other workers. Blockade of protein synthesis by the parasite following concentration of the compounds in infected erythrocytes appears to be the primary lesion that they cause.

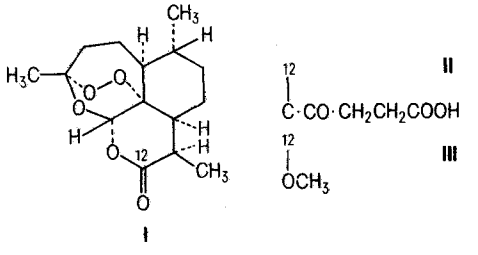

Figure 2. **I**, Artemisinine; **II**, artesunate; **III**, artemether.

Other blood schizontocides

While various other compounds have been described in recent literature, very few outside the WRAIR programme[5] have passed their preclinical trial stage. New developments were recently reviewed[19, 41] and have been further detailed by various authors[31]. Among the newer compounds hydroxypiperaquine (fig. 3, **I**), pyronaridine (fig. 3, **II**) and halofantrene (fig. 1, **II**) are known to be in clinical trial.

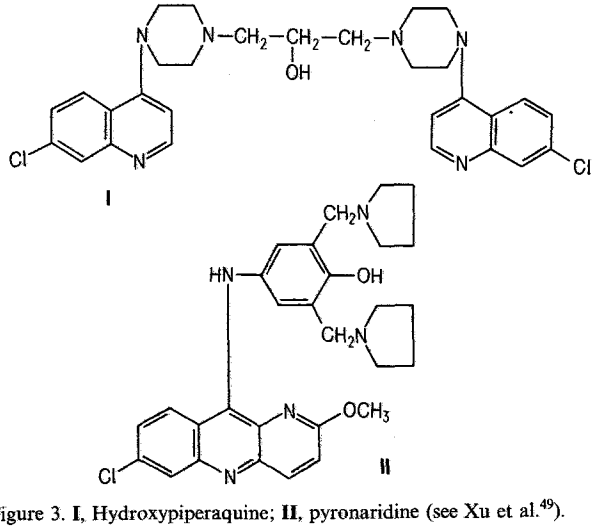

Figure 3. **I**, Hydroxypiperaquine; **II**, pyronaridine (see Xu et al.[49]).

Unfortunately the potentiating combination of pyrimethamine with sulfadoxine (Fansidar) which proved so effective in the treatment of multiple-resistant *P. falciparum* infections for the past 15 years has now lost its edge with the development of an increasing proportion of Fansidar-resistant strains of this parasite in Southeast Asia, South America and, recently, East Africa. Nevertheless the marked difference between the speed with which resistance to this combination has appeared in the field with the rate at which resistance appeared to pyrimethamine when it was used alone (e.g. a matter of months only in Ghana)[6] is most striking and should be remembered by future policy makers.

Tissue schizontocides

Combined in this category are both those compounds that function as causal prophylactics (sensu lato) and hypnozoitocides. The review by Davidson et al.[10] and subsequent analysis of the problem by a group of WHO experts[42] made it clear that, of the numerous chemical groups which had displayed clearcut prophylactic activity in various rodent malaria models in vivo, only 8-aminoquinolines had proved to be hypnozoitocidal when tested in the *P. cynomolgi*-rhesus model. Recent studies by workers in Paris and London[4] suggest that primaquine at least may exert an effect both on the pre-erythrocytic schizont and its hepatic parenchymal host cell. Other investigators are exploring the metabolites and pharmacokinetics of the 8-aminoquinolines in the hope of detecting non-toxic but antimalarial metabolites.

Several new 8-aminoquinolines that are being evaluated as possible replacements for primaquine show promise in having a strong blood schizontocidal as well as tissue schizontocidal effect. Such a compound is WR 225 448 (fig. 4, **I**)[10,19].

A further compound, floxacrine (fig. 4, **II**) showed great interest in having both blood and tissue schizontocidal action in rodent models[34] but it too did not have hypnozoitocidal action against *P. cynomolgi* at the maximum tolerated dose, and was dropped when it showed unexpected, but prohibitive toxicity in animal studies[37].

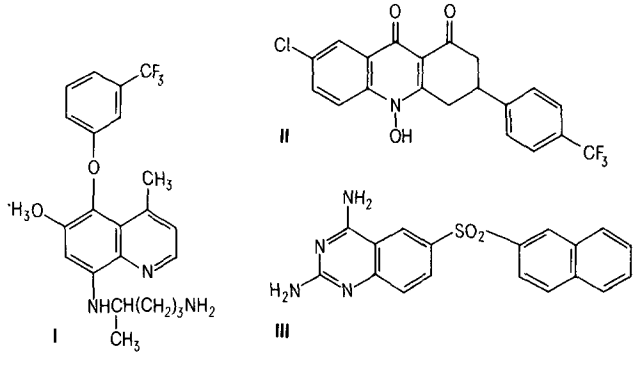

Figure 4. **I**, WR 225448; **II**, floxacrine; **III**, WR 158122.

Repository preparations

While I do not believe that repository preparations offer any serious advantage for chemoprophylaxis or therapy in the individual, they clearly do in 2 indications for communal use, i.e. when 'collective drug protection' is indicated (and here either a good causal prophylactic or suppressive drug is needed), or for the radical cure of vivax infections when adequate supervision of prolonged courses (e.g. of primaquine) is impractical. Trouet et al.[45] described several series of primaquine derivatives including peptidic prodrugs with which they were able to extend the half-life and reduce the toxicity of the parent compound in rodent malaria models, but only to a modest degree. Other workers have synthesised prodrugs based on other procedures but details of their data are not yet available.

Attention has been drawn to the screening of compounds for inherently long biological half-lives[39]. Relatively long half-lives in this series appeared to be linked mainly to poor solubility and the formation of local depots at injection sites.

Following the lead of Thompson and his associates[43] who produced promising, but irritant injectable formulations containing poorly soluble salts of cycloguanil and dapsone, Wise et al.[48] studied a series of mixtures of the compound WR 158122 (fig. 4, **III**), a poorly soluble quinazoline, alone or with sulphadiazine, incorporated in biodegradable polymers based on lactic and glycolic acids. They experienced considerable difficulty in obtaining the parallel release rates of the two compounds which would have been necessary if each compound was to 'protect' the other i.e. achieve optimal potentiation of action while minimising the rate at which the parasites could become resistant to them. Subsequently Judge et al.[20] obtained very prolonged activity in rodent malaria models with subcutaneous implants containing pyrimethamine and sulphadiazine in a different type of biodegradable polymer, a cross-linked polydihydropyran, 20 and 37 weeks respectively. No new repository formulations have yet reached the stage of clinical trial. One major problem here is the necessity to use appropriate combinations of drugs, preferably potentiating pairs, to minimize the risk of selecting resistant parasites, and this in turn greatly complicates the preclinical efficacy and safety testing that must be performed, not to mention the cost of developing such preparations.

The role of drug combinations in preventing drug resistance

One theme that has run throughout this chapter has been the danger that parasites resistant to virtually any drug will in time be selected, and that the more extensively a drug is deployed, the more rapidly will emerge resistant organisms. In the case of antifols the risk is so great that it is now considered unethical if not suicidal to administer an antifol alone, be it for the treatment of malaria or a bacterial infection. The example of Fansidar quoted above does show the value of an appropriately selected drug combination in protecting the individual components against resistance. It is essential to stress the word *appropriate* since this factor must be determined by preliminary study in a good experimental model before deciding on which compounds to associate. Thus Merkli et al.[24] have shown that the triple com-

bination of mefloquine, pyrimethamine and sulfadoxine* is extremely effective in delaying the emergence of resistance to any of the three in malaria infected mice, while we have confirmed that this holds for mefloquine even in a strain already resistant to the other two components[32]. On the basis of painful past experience with older drugs used alone, or with chloroquine administered with pyrimethamine, and in the face both of established Fansidar and emerging quinine resistance in *P. falciparum*, extensive studies are now being made under WHO TDR auspices of this triple combination for use in man. While mefloquine will shortly be made available for the treatment of individuals with acute, chloroquine-resistant falciparum malaria, it is feared that the release of this compound *alone* for prophylaxis will rapidly be followed by its misuse and subsequently by the appearance of resistance even to this valuable new drug. It is hoped and anticipated that the deployment of the triple combination will extend the life of mefloquine and Fansidar for sufficient time for the next generation of antimalarials to be developed. It takes at least 10 years development from first discovery to introduction for a new compound to become available for use in man.

Meanwhile other means are being sought to protect both the individual and the community against malaria. Other chapters in this review outline progress that is being made in developing new means of vector control, and high hopes are currently being held out for the production of safe vaccines with which to protect man. Both approaches however pose many problems and many hurdles will have to be crossed before new weapons in the fight against malaria become available. When they do, and this is particularly important in the case of vaccines as well as of drugs, it is vital to keep in mind the distinction between protecting the individual and protecting the community. We have made this mistake in the past. Let us learn from our mistakes.

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Malaria prophylaxis in travellers: the current position

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Summary. Malaria prevention is a main challenge for physicians, nurses, health officers and tour operators. The attack rate of malaria in travellers is 1-10/10,000 departures, and the case fatality rate of imported malaria is around 0.5/100. Travellers should be informed about the risk they are going to take, how to protect against mosquito bites, about the antimalarials they will have to take and about what to do when a malaria breakthrough should occur.

The 4-aminoquinolines (chloroquine, amodiaquine) remain the drug of choice for the prevention of *Plasmodium vivax* and of sensitive *P. falciparum* infections. The problem is to find an effective and safe drug combination for travellers to areas where *P. falciparum* is either resistant to chloroquine, to Fansidar (the combination of pyrimethamine plus sulfadoxine) or to both. These travellers will probably best be protected by an individually tailored drug combination, which includes amodiaquine or mefloquine as baseline drugs, and a supplementation with Fansidar, Maloprim (the combination of pyrimethamine with dapsone), paludrine or an antibiotic.

Key words. Malaria; prophylaxis, malaria; Plasmodia; mosquito bite, protection against; antimalarial drugs.

Introduction

Malaria is perhaps the most important parasitic infection worldwide. It covers vast areas in Africa, Asia and Latin America (fig. 1). According to incomplete reportings to WHO there were 13 million malaria cases in 1981[32]. One expert estimates that the disease affects more than 200 million people each year[24]. Of the 179 states where there is a potential for malaria transmis-